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(71) Applicant *(for all designated States except US)*: **ANTISOMA RESEARCH LIMITED** [GB/GB]; West Africa House, Hanger Lane, Ealing, London W5 3QR (GB).

(72) Inventor; and

(75) Inventor/Applicant *(for US only)*: **YOUNG, Robert, James** [GB/GB]; Antisoma Research Limited, West Africa House, Hanger Lane, Ealing, London W5 3QR (GB).

(74) Agent: **THOMAS, Philip, J., D.**; Eric Potter Clarkson, Park View House, 58 The Ropewalk, Nottingham NG1 5DD (GB).

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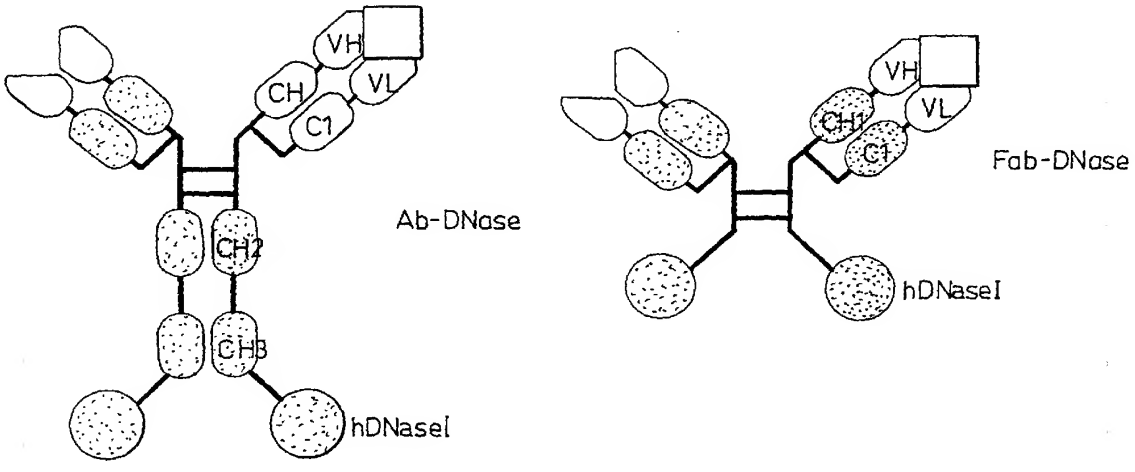

(57) Abstract: A compound comprising a target cell-specific portion and a cytotoxic portion characterised in that the target cell-specific portion comprises a humanised monoclonal antibody having specificity for polymorphic epithelial mucin (PEM), or an antigen binding fragment thereof, and the cytotoxic portion has endonucleolytic activity. Preferably, the target cell-specific portion comprises a humanised HMFG-1 antibody or an antigen binding fragment thereof. Advantageously, the cytotoxic portion is at least the catalytically active portion of a DNA endonuclease, e.g. a human DNA endonuclease I. The invention further provides nucleic acids encoding the compounds of the invention, and the use of such compounds in medicine, e.g. in the treatment of cancer.

WO 01/74905 A1

CLAIMS

1. A compound comprising a target cell-specific portion and a cytotoxic portion characterised in that:

   (i) the target cell-specific portion comprises an humanised monoclonal antibody having specificity for polymorphic epithelial mucin (PEM), or an antigen binding fragment thereof; and

   (ii) the cytotoxic portion has endonucleolytic activity.

2. A compound according to Claim 1 wherein the target cell-specific portion comprises an humanised HMFG-1 antibody or an antigen binding fragment thereof.

3. A compound according to Claim 2 wherein the target cell-specific portion is an humanised HMFG-1 antibody.

4. A compound according to Claim 1 or 2 wherein the target cell-specific portion comprises an antigen binding fragment of the humanised antibody selected from the group consisting of Fab-like molecules, such as Fab and $F(ab')_2$, Fv molecules, disulphide-linked Fv molecules, ScFv molecules and single domain antibodies (dAbs).

5. A compound according to Claim 4 wherein the target cell-specific portion comprises a Fab molecule.

6. A compound according to Claim 4 wherein the target cell-specific

portion comprises a $F(ab')_2$ molecule.

7. A compound according to Claim 1 wherein the target cell-specific portion comprises an amino acid sequence encoded by at least part of one or both of the nucleotide sequences of Figure 3(a) and (d).

8. A compound according to Claim 7 wherein the target cell-specific portion comprises an amino acid sequence encoded by the nucleotide sequence of Figure 3(a) and an amino acid sequence encoded by the nucleotide sequence of Figure 3(d).

9. A compound according to any one of Claims 1 to 8 wherein the cytotoxic portion has DNA endonucleolytic activity.

10. A compound according to Claim 9 wherein the cytotoxic portion is at least the catalytically active portion of a DNA endonuclease.

11. A compound according to Claim 10 wherein the endonuclease is a mammalian deoxyribonuclease I.

12. A compound according to Claim 11 wherein the endonuclease is a human deoxyribonuclease I.

13. A compound according to Claim 1 wherein the endonuclease is a restriction endonuclease.

14. A compound according to Claim 10 wherein the cytotoxic portion comprises the amino acid sequence shown in Figure 2(a) or (b).

15. A compound according to any one of Claims 1 to 14 wherein a nuclear localization signal is incorporated.

16. A compound according to Claim 15 wherein the nuclear localization signal comprises the sequence PKKKRKV.

17. A compound according to any one of Claims 1 to 16 wherein the target cell-specific portion and the cytotoxic portion are fused.

18. A compound according to Claim 17 wherein the target cell-specific portion and the cytotoxic portion are separated by a linker sequence.

19. A compound according to Claim 18 wherein the linker sequence is or comprises GG or GSGG.

20. A compound according to any one of Claims 1 to 19 wherein the compound comprises all or part of the amino acid sequence as shown in Figure 3(c) together with all or part of an amino acid sequence selected from the group consisting of amino acid sequences as shown in Figures 5(d), 6(d), 7(b), 8(b), 9(b), 10(b), 11(b), 12(b), 13(d), 14(d), 15(d), 16(c), 17(d), 18(d) and 19(d).

21. A compound according to Claim 20 wherein the compound comprises an amino acid sequence as shown in Figure 3(c) and an amino acid sequence as shown in Figure 7(b).

22. A compound according to Claim 20 wherein the compound comprises

an amino acid sequence as shown in Figure 3(c) and an amino acid sequence as shown in Figure 14(d).

23. A nucleic acid molecule encoding a compound as defined in any one of Claims 1 to 22.

24. A nucleic acid molecule according to Claim 23 wherein the molecule comprises all or part of the nucleotide sequence as shown in Figure 3(a or b) together with all or part of a nucleotide sequence selected from the group consisting of nucleotide sequences as shown in Figures 5(a, b and c), 6(a, b and c), 7(a), 8(a), 9(a), 10(a), 11(a), 12(a), 13(a, b and c), 14(a, b and c), 15(a, b and c), 16(a and b), 17(a, b and c), 18(a, b and c) and 19(a, b and c).

25. A nucleic acid molecule according to Claim 24 wherein the molecule comprises a nucleotide sequence as shown in Figure 3(b) and a nucleotide sequence as shown in Figure 7(a).

25. A nucleic acid molecule according to Claim 24 wherein the molecule comprises a nucleotide sequence as shown in Figure 3(b) and a nucleotide sequence as shown in Figure 14(c).

26. A nucleic acid molecule according to any one of Claims 23 to 25 wherein the molecule further comprises a Kozak consensus ribosome-binding site.

27. A vector comprising a nucleic acid molecule according to any one of Claims 23 to 26.

28. A host cell comprising a vector according to Claim 27.

29. A pharmaceutical composition comprising a compound according to any one of Claims 1 to 22 and a pharmaceutically acceptable carrier.

30. A compound according to any one of Claims 1 to 22 for use in medicine.

31. Use of a compound according to any one of Claims 1 to 22 in the preparation of a medicament for treating a mammal having said target cells to be destroyed.

32. A method of treating a mammal having target cells to be destroyed, the method comprising administering a compound according to any one of Claims 1 to 22 to said mammal.

33. A use according to Claim 31 or a method according to Claim 32 wherein the mammal is a human.

34. A use according to Claim 31 or a method according to Claim 32 wherein the target cells to be destroyed are cancer cells.

35. A use or a method according to Claim 34 wherein the cancer cells are epithelial cancer cells.

36. A use or a method according to Claim 35 wherein the cancer cells are ovarian, gastric, colorectal and/or pancreatic cancer cells.

37. A use or a method according to Claim 36 wherein the cancer cells are ovarian cancer cells.

38. A compound substantially as described herein, preferably with reference to one or more of the accompanying figures.

COMPOUNDS FOR TARGETING

The present invention relates to cytotoxic compounds that have a high avidity for, and can be targeted to, selected cells. Specifically, the invention provides compounds comprising a cytotoxic portion having DNA endonucleolytic activity and a target-cell specific portion having specificity for human polymorphic epithelial mucin (PEM).

Background

The cell-specific targeting of compounds that are directly, or indirectly, cytotoxic has been proposed as a way to combat diseases such as cancer. Bagshawe and his co-workers have disclosed (Bagshawe (1987) *Br. J. Cancer* **56**, 531; Bagshawe *et al* (1988) *Br. J. Cancer* **58**, 700; WO 88/07378) conjugated compounds comprising an antibody or part thereof and an enzyme, the antibody being specific to tumour cell antigens and the enzyme acting to convert an innocuous pro-drug into a cytotoxic compound. The cytotoxic compounds were alkylating agents, *e.g.* a benzoic acid mustard released from *para*-N-bis(2-chloroethyl)aminobenzoyl glutamic acid by the action of *Pseudomonas sp.* CPG2 enzyme.

An alternative system using different pro-drugs has been disclosed (WO 91/11201) by Epenetos and co-workers. The cytotoxic compounds were cyanogenic monosaccharides or disaccharides, such as the plant compound amygdalin, which release cyanide upon the action of a β-glucosidase and hydroxynitrile lyase.

In a further alternative system, the use of antibody-enzyme conjugates containing the enzyme alkaline phosphatase in conjunction with the pro-drug etoposide 4'-phosphate or 7-(2'-aminoethyl phosphate)mitomycin or a combination thereof have been disclosed (EP 0 302 473; Senter *et al* (1988) *Proc. Natl. Acad. Sci. USA* **85**, 4842).

Rybak and co-workers have disclosed (Rybak *et al* (1991) *J. Biol. Chem.* **266**, 21202; WO 91/16069) the cytotoxic potential of a monomeric pancreatic ribonuclease when injected directly into *Xenopus* oocytes and the cytotoxic potential of monomeric RNase coupled to human transferrin or antibodies directed against the transferrin receptor. The monomeric RNase hybrid proteins were cytotoxic to human erythroleukaemia cells *in vitro*.

Other approaches are the *in vivo* application of streptavidin conjugated antibodies followed, after an appropriate period, by radioactive biotin (Hnatowich *et al* (1988) *J. Nucl. Med.* **29**, 1428-1434), or injection of a biotinylated mAb followed by radioactive streptavidin (Paganelli *et al* (1990) *Int. J. Cancer* **45**, 1184-1189). A pilot radioimmunolocalisation study in non-small cell lung carcinomas was conducted with encouraging results (Kalofonos *et al* (1990) *J. Nucl. Med.* **31**, 1791-1796).

Apart from these examples, it is rather more common to see biotinylated antibodies and streptavidin-enzyme conjugates, which are used in enzyme-linked immunosorbent assays.

These previous systems have used relatively large antibody-enzyme,

antibody-streptavidin or antibody-biotin conjugates and may comprise portions of non-mammalian origin which are highly immunoreactive.

We have now devised improved compounds for targeting cells to be destroyed.

Summary of Invention

A first aspect of the invention provides a compound comprising a target cell-specific portion and a cytotoxic portion characterised in that the target cell-specific portion comprises an humanised monoclonal antibody having specificity for polymorphic epithelial mucin (PEM), or an antigen binding fragment thereof, and the cytotoxic portion has endonucleolytic activity.

By "target cell specific" portion we mean the portion of the compound which comprises one or more binding sites which recognise and bind to polymorphic epithelial mucin (PEM) on the target cell. Upon contact with the target cell, the target cell specific portion is preferably internalised along with the cytotoxic portion. Such internalisation results in the cytotoxic portion being delivered to the cell cytosol, where it has access to the cell's nucleic acid molecules.

The target cell-specific portion of the compounds of the invention comprises an humanised monoclonal antibody having specificity for polymorphic epithelial mucin (PEM), or an antigen binding fragment thereof.

Polymorphic epithelial mucin, or PEM, is a component of the human milk

fat globule. PEM is expressed by cells in several body tissues and is also found in urine. Significantly, PEM is known to be expressed in epithelial cancer cells, notably in ovarian, gastric, colorectal and pancreatic cancer cells.

Monoclonal antibodies which will bind to PEM are already known, but in any case, with today's techniques in relation to monoclonal antibody technology, antibodies can be prepared to most antigens. The antigen-specific portion may be a whole antibody, a part of an antibody (for example a Fab or $F(ab')_2$ fragment), a synthetic antibody fragment (for example a single chain Fv fragment [ScFv]), or a peptide/peptidomimetic or similar. Suitable monoclonal antibodies to selected antigens may be prepared by known techniques, for example those disclosed in *"Monoclonal Antibodies: A manual of techniques"*, H Zola (CRC Press, 1988) and in *"Monoclonal Hybridoma Antibodies: Techniques and Applications"*, J G R Hurrell (CRC Press, 1982) and *Antibody Engineering, A Practical Approach*, McCafferty, J. et al, ed. (IRL Pres, 1996).

By 'humanised monoclonal antibody' we include monoclonal antibodies having at least one chain wherein the framework regions are predominantly derived from a first, acceptor monoclonal antibody of human origin and at least one complementarity-determining region (CDR) is derived from a second, donor monoclonal antibody having specificity for PEM. The donor monoclonal antibody may be of human or non-human origin, for example it may be a murine monoclonal antibody.

Preferably, both chains of the humanised monoclonal antibody comprise

CDRs grafted from a donor monoclonal antibody having specificity for PEM.

Advantageously, the CDR-grafted (*i.e.* humanised) chain comprises two or all three CDRs derived from a donor antibody having specificity for PEM.

Conveniently, the humanised monoclonal antibody comprises only human framework residues and CDRs from a donor antibody having specificity for PEM.

However, it will be appreciated by those skilled in the art that in order to maintain and optimise the specificity of the humanised antibody it may be necessary to alter one or more residues in the framework regions such that they correspond to equivalent residues in the donor antibody.

Conveniently, the framework regions of the humanised antibody are derived from an human IgG monoclonal antibody.

Methods of making humanised monoclonal antibodies are well-known in the art, for example see Jones *et al.* (1986) *Nature* **321**:522-525, Riechmann *et al.* (1988) *Nature* **332**:323-327, Verhoeyen *et al.* (1988) *Science* **239**:1534-1536 and EP 239 400 (to Winter).

In a preferred embodiment of the first aspect of the invention, the target cell-specific portion comprises an humanised HMFG-1 monoclonal antibody or an antigen binding fragment thereof.

HMFG antibodies are raised against human milk fat globule (HMFG), in a delipidated state (see Taylor-Papadimiriou *et al.*, 1981, *Int. J. Cancer* **28**:17-21 and Gendler *et al.*, 1988, *J. Biol. Chem.* **236**:1282-12823). HMFG-1 monoclonal antibodies bind to a particular component of HMFG, namely polymorphic epithelial mucin (PEM). Binding is thought to involve the amino acid sequence APDTR within the twenty amino acid tandem repeats of the *muc-1* gene product.

Exemplary humanised HMFG-1 antibodies are disclosed in WO 92/04380.

Advantageously, the target cell-specific portion is an humanised HMFG-1 monoclonal antibody.

In a preferred embodiment of the first aspect of the invention, the target cell-specific portion comprises a fragment of an humanised monoclonal antibody having specificity for polymorphic epithelial mucin (PEM), said fragment retaining the antigen binding properties of the parent antibody.

The variable heavy (V_H) and variable light (V_L) domains of the antibody are involved in antigen recognition, a fact first recognised by early protease digestion experiments. Further confirmation was found by "humanisation" of rodent antibodies. Variable domains of rodent origin may be fused to constant domains of human origin such that the resultant antibody retains the antigenic specificity of the rodent parented antibody (Morrison *et al* (1984) *Proc. Natl. Acad. Sci. USA* **81**, 6851-6855).

That antigenic specificity is conferred by variable domains and is independent of the constant domains is known from experiments involving

the bacterial expression of antibody fragments, all containing one or more variable domains. These molecules include Fab-like molecules (Better *et al* (1988) *Science* **240**, 1041); Fv molecules (Skerra *et al* (1988) *Science* **240**, 1038); disulphide-linked Fv molecules (Young *et al.*, 1995, *FEBS Lett.* **377**:135-139); single-chain Fv (ScFv) molecules where the V_H and V_L partner domains are linked via a flexible oligopeptide (Bird *et al* (1988) *Science* **242**, 423; Huston *et al* (1988) *Proc. Natl. Acad. Sci. USA* **85**, 5879) and single domain antibodies (dAbs) comprising isolated V domains (Ward *et al* (1989) *Nature* **341**, 544). A general review of the techniques involved in the synthesis of antibody fragments which retain their specific binding sites is to be found in Winter & Milstein (1991) *Nature* **349**, 293-299.

By "ScFv molecules" we mean molecules wherein the V_H and V_L partner domains are linked via a flexible oligopeptide.

Chimaeric antibodies are discussed by Neuberger *et al* (1988, *8th International Biotechnology Symposium* Part 2, 792-799).

The advantages of using antibody fragments, rather than whole antibodies, are several-fold. The smaller size of the fragments allows for rapid clearance, and may lead to improved tumour to non-tumour ratios. Fab, Fv, ScFv, disulphide Fv and dAb antibody fragments can all be expressed in and secreted from bacteria, such as *E. coli*, or eukaryotic expression systems such as Yeast or mammalian systems, thus allowing the facile production of large amounts of the said fragments.

Whole antibodies, and $F(ab')_2$ fragments are "bivalent". By "bivalent" we

mean that the said antibodies and $F(ab')_2$ fragments have two antigen combining sites. In contrast, Fab, Fv, ScFv, disulphide Fv and dAb fragments are monovalent, having only one antigen combining site.

Preferably, the target cell-specific portion of the compounds of the invention comprises an antigen binding fragment of the humanised antibody selected from the group consisting of Fab-like molecules, such as Fab and $F(ab')_2$, Fv molecules, disulphide-linked Fv molecules, ScFv molecules and single domain antibodies (dAbs).

More preferably, the target cell-specific portion comprises a Fab molecule or a $F(ab')_2$ molecule.

Yet more preferably, the target cell-specific portion comprises an amino acid sequence encoded by at last part of one or both of the nucleotide sequences of Figure 3(a) and (d).

Most preferably, the target cell-specific portion comprises an amino acid sequence encoded by the nucleotide sequence of Figure 3(a) and an amino acid sequence encoded by the nucleotide sequence of Figure 3(d).

Preferably, the target cell-specific portion recognises the target cell with high avidity.

By "high avidity" we mean that the target cell-specific portion recognises the target cell with a binding constant of at least $K_d = 10^{-6}$ M, preferably at least $K_d = 10^{-9}$ M, suitably $K_d = 10^{-10}$ M, more suitably $K_d = 10^{-11}$ M, yet more suitably still $K_d = 10^{-12}$ M, and more preferably $K_d = 10^{-15}$M or

even $K_d = 10^{-18}$ M.

Preferably, the target cell-specific portion comprises an antigen binding fragment of an humanised HMFG-1 monoclonal antibody, *e.g.* an Fab or F(ab')$_2$ fragment thereof, wherein a hinge region contains a mutation (*i.e.* wherein the hinge is a variant or hybrid of a naturally occurring hinge). More preferably, the variant hinge comprises the amino acid sequence CCVECPPCPAPE.

By 'cytotoxic portion' we mean a portion having endonucleolytic activity which is toxic to the cell if it is to reach, and preferably enter said cell.

In a preferred embodiment of the first aspect of the invention, the cytotoxic portion has DNA endonucleolytic activity.

Advantageously, the cytotoxic portion is at least the catalytically active portion of a DNA endonuclease.

Examples of known DNA endonucleases include bovine DNase I (see Worrall and Conolly, 1990, *J. Biol. Chem.* **265**:21889-21895). Human pancreatic DNase I has also been cloned (see Shak *et al.*, 1990, *Proc. Natl. Acad. Sci. USA* **87**:9188-9192 and Hubbard *et al.*, 1992, *New Eng. J. Med.* **326**:812-815).

Preferably, the endonuclease is a mammalian deoxyribonuclease I.

More preferably, the endonuclease is a human deoxyribonuclease I.

Most preferably, the cytotoxic portion comprises the amino acid sequence shown in Figure 2(a) or 2(b).

Preferably, the cytotoxic portion of the compound of the invention is capable of oligomerisation, *e.g.* dimerisation. Attachment of the target-cell specific portion to a cytotoxic portion capable of oligomerisation provides a method for increasing the number of binding sites to the target cell. For example, if the target cell-specific portion is joined to a portion capable of forming a dimer then the number of target cell-specific binding sites is two; if the target cell-specific portion is joined to a portion capable of forming a tetramer then the number of target cell-specific binding sites is four. The number of target cell-specific binding sites is greater than one and the compounds may therefore have a greater avidity for the target cell than do compounds which only have one target cell-specific binding site.

It is preferable for the cytotoxic portion of the compound of the invention capable of oligomerisation to contain no interchain disulphide bonds nor intrachain disulphide bonds; to be well characterised; to be non-toxic; to be stable; to be amenable to preparation in a form suitable for pre-clinical or clinical use or be in pre-clinical or clinical use; and for the subunit monomers to have a high affinity for each other, that is they contain one or more subunit binding sites.

Advantageously, the cytotoxic portion is of mammalian, preferably human, origin. The use of the said mammalian proteins as the cytotoxic portion of the compound of the invention is advantageous since such compounds are less likely to give rise to undesirable immune reactions.

It will be appreciated by those skilled in the art that the cytotoxic portion may be a variant of a naturally occurring endonuclease.

By "a variant" we include cytotoxic portions comprising of a naturally occurring endonuclease wherein there have been amino acid insertions, deletions or substitutions, either conservative or non-conservative, such that the changes do not substantially reduce the endonuclease activity of the variant compared to that of the naturally occurring endonuclease. For example, the variant may have increased activity compared to the naturally occurring endonuclease

Such variants may be made using methods of protein engineering and site-directed mutagenesis commonly known in the art (for example, see Sambrook *et al.*, 1989, *Molecular cloning: A Laboratory Manual*, 2nd edition, Cold Spring Harbor Laboratory Press, NY, USA).

In an alternative embodiment, the endonuclease is a restriction endonuclease, such as a microbial type II restriction endonuclease. Exemplary type II restriction endonucleases include *Bam*HI, *Hind*III, *Msp*I, *Sau*3AI, *Hinf*I, *Not*I and *Eco*RI.

In another preferred embodiment of the first aspect of the invention, a nuclear localization signal is incorporated into the compound.

Preferably, the nuclear localization signal (NLS) comprises a nuclear localization signal from the SV40 large T antigen (Kalderon *et al.*, 1984, *Cell* **39**:499-509), and specifically the amino acid sequence PKKKRKV.

Inclusion of a nuclear localization signal encourages the compound of the invention to gain access to the chromosomal DNA during the periods of the cell cycle when the nuclear membrane is intact, since the nuclear pores are permeable to large molecules incorporating said nuclear localization signal.

In a further preferred embodiment of the first aspect of the invention, the target cell-specific portion and the cytotoxic portion are fused to create a fusion compound.

By "fusion compound" we include a compound comprising one or more functionally distinct portions, wherein the distinct portions are contained within a single polypeptide chain produced by recombinant DNA techniques. For example, the compound may comprise a whole antibody wherein the heavy chain is fused to human DNase I. Alternatively, the compound may comprise an Fab or $F(ab')_2$ fragment of an antibody wherein the truncated heavy chain (*i.e.* the Fd chain) is fused to human DNase I.

Preferably, the target-cell specific and the cytotoxic portion of the fusion compound of the invention separated by a linker sequence, for example to allow greater flexibility of the portions relative to one another.

More preferably, the linker sequence comprises a GG dipeptide.

Most preferably the linker sequence is or comprises GG or GSGG.

Alternatively, the target-cell specific and the cytotoxic portion of the

compound of the invention are separate moieties linked together by any of the conventional ways of cross-linking polypeptides, such as those generally described in O'Sullivan *et al Anal. Biochem.* (1979) **100**, 100-108. For example, the antibody portion may be enriched with thiol groups and the enzyme portion reacted with a bifunctional agent capable of reacting with those thiol groups, for example the N-hydroxysuccinimide ester of iodoacetic acid (NHIA) or N-succinimidyl-3-(2-pyridyldithio)propionate (SPDP). Amide and thioether bonds, for example achieved with m-maleimidobenzoyl-N-hydroxysuccinimide ester, are generally more stable *in vivo* than disulphide bonds.

In a preferred embodiment of the first aspect of the invention, the compound comprises all or part of the amino acid sequence as shown in Figure 3(c) (*i.e.* an HMFG-1 light chain) together with all or part of an amino acid sequence selected from the group consisting of amino acid sequences as shown in Figures 5(d), 6(d), 7(b), 8(b), 9(b), 10(b), 11(b), 12(b), 13(d), 14(d), 15(d), 16(c), 17(d), 18(d) and 19(d) (*i.e.* an HMFG-1 heavy or Fd chain/DNase fusion).

Advantageously, the compound is a whole HMFG-1 antibody/human DNase I fusion compound comprising an amino acid sequence as shown in Figure 3(c) and an amino acid sequence as shown in Figure 7(b). Preferably, the compound is a tetrameric compound comprising two HMFG-1 light chains and two HMFG-1 heavy chain /DNase I fusions.

Conveniently, the compound comprises an amino acid sequence as shown in Figure 3(c) and an amino acid sequence as shown in Figure 14(d).

Preferably, the compound comprises one of the pairs of amino acid sequences defined above wherein the leader sequence of each amino acid (the first 19 amino acids of the sequences shown in each figure) is removed. It will be appreciated by persons skilled in the art that the compounds of the invention may also comprise variants of such amino acid sequences.

Suitably, the compound is a tetrameric compound comprising two HMFG-1 light chains and two HMFG-1 Fd chain /DNase I fusions. More preferably, the compound is a dimeric compound comprising one HMFG-1 light chain and one HMFG-1 Fd chain /DNase I fusion.

A second aspect of the invention provides a nucleic acid molecule encoding a compound according to the first aspect of the invention, or a target cell-specific portion or cytotoxic portion thereof.

By "nucleic acid molecule" we include DNA, cDNA and mRNA molecules.

In a preferred embodiment of the second aspect of the invention, the nucleic acid molecule comprises all or part of the nucleotide sequence as shown in Figure 3(a or b) (*i.e.* encoding an HMFG-1 light chain) together with all or part of a nucleotide sequence selected from the group consisting of nucleotide sequences as shown in Figures 5(a, b and c), 6(a, b and c), 7(a), 8(a), 9(a), 10(a), 11(a), 12(a), 13(a, b and c), 14(a, b and c), 15(a, b and c), 16(a and b), 17(a, b and c), 18(a, b and c) and 19(a, b and c) (*i.e.* encoding an HMFG-1 heavy or Fd chain/DNase fusion).

Advantageously, the nucleic acid molecule comprising a nucleotide sequence as shown in Figure 3(b) and a nucleotide sequence as shown in Figure 7(a).

Conveniently, the compound comprises a nucleotide sequence as shown in Figure 3(b) and a nucleotide sequence as shown in Figure 14(c).

Alternatively, the nucleic acid molecule comprises nucleotide sequences that are degenerate sequences of those nucleotide sequences identified above (*i.e.* which encode the same amino acid sequence).

A further aspect of the present invention provides a method of making a compound according to the first aspect of the invention, said method comprising expressing one or more nucleic acid molecules according to the second aspect of the invention in a host cell and isolating the compound therefrom.

It is preferable that the two portions of the compound of the invention are produced as a fusion compound by recombinant DNA techniques, whereby a length of DNA comprises respective regions encoding the two portions of the compound of the invention either adjacent one another or separated by a region encoding a linker peptide which does not destroy the desired properties of the compound. The benefits in making the compound of the invention using recombinant DNA techniques are several fold. Firstly, it enables a high degree of precision with which the two portions of the compound can be joined together. Secondly, the construction of compounds which are "hetero-oligomeric" can be controlled by the expression of the different recombinant DNA molecules

encoding each of the different type of subunit of the "hetero-oligomer" in the same host cell.

By "hetero-oligomer" we mean those compounds in which two or more different cell-specific portions are joined to either the same or to different subunits which are capable of oligomerisation. The expression, in the same host cell of two compounds, of A and B, each with different target cell specific portions but with a common second portion capable of oligomerisation will result in a mixed population of compounds. For example, if the common second portion is capable of dimerisation, three potential compounds will be produced: A_2, AB and B_2, in a ratio of 1:2:1, respectively.

The separation of the desired compound with each of the different cell specific portions, that is AB, can be achieved by two step affinity chromatography.

Application of the mixture of compounds to an affinity column specific for A will result in the binding of A_2 and AB. These compounds are eluted from this first column, and then applied to an affinity column specific for B. This will result in AB, but not A_2, being bound to the column. Finally, the desired product AB, can be eluted.

Of course, the order in which the affinity columns are used is not important.

The same principle of separating those compounds with two or more different binding sites can be applied to the purification of the desired

compounds from mixtures of other hetero-oligomers.

Conceivably, the two portions of the compound may overlap wholly or partly.

Preferably, the compound is a multimeric compound such as a whole antibody/DNase fusion comprising two light chains and two heavy chains (H_2L_2), a $F(ab')_2$ fusion comprising two light chains and two truncated heavy chains (Fd_2L_2), or a Fab fusion comprising one light chain and one truncated heavy chain (FdL).

The nucleic acid may be expressed in a suitable host to produce a polypeptide comprising the compound of the invention. Thus, the nucleic acid encoding the compound of the invention or a portion thereof may be used in accordance with known techniques, appropriately modified in view of the teachings contained herein, to construct an expression vector, which is then used to transform an appropriate host cell for the expression and production of the polypeptide of the invention. Such techniques include those disclosed in US Patent Nos. 4,440,859 issued 3 April 1984 to Rutter *et al*, 4,530,901 issued 23 July 1985 to Weissman, 4,582,800 issued 15 April 1986 to Crowl, 4,677,063 issued 30 June 1987 to Mark *et al*, 4,678,751 issued 7 July 1987 to Goeddel, 4,704,362 issued 3 November 1987 to Itakura *et al*, 4,710,463 issued 1 December 1987 to Murray, 4,757,006 issued 12 July 1988 to Toole, Jr. *et al*, 4,766,075 issued 23 August 1988 to Goeddel *et al* and 4,810,648 issued 7 March 1989 to Stalker, all of which are incorporated herein by reference.

Where the compound of the invention is multimeric, the constituent chains

may be encoded by a single nucleic acid molecule or separate nucleic acid molecule (expressed in a common host cell or in different host cells and assembled *in vitro*).

The nucleic acid encoding the compound of the invention or a portion thereof may be joined to a wide variety of other nucleic acid sequences for introduction into an appropriate host. The companion nucleic acid will depend upon the nature of the host, the manner of the introduction of the nucleic acid into the host, and whether episomal maintenance or integration is desired.

It will be appreciated that in order to prevent expression of the cytotoxic portion of the compound of the invention from killing the host cells in which it is expressed, it may be necessary to link the nucleic acid of the second aspect of the invention to a signal sequence capable of directing secretion of the expressed compound (or portion) out of the host cell. Signal sequences will be selected according to the type of host cell used. Exemplary signal sequences include the *ompA* signal sequence (for example, see Takahara *et al.*,1985, *J. Biol. Chem.* **260(5)**:2670-2674).

Generally, the nucleic acid is inserted into an expression vector, such as a plasmid, in proper orientation and correct reading frame for expression. If necessary, the nucleic acid may be linked to the appropriate transcriptional and translational regulatory control nucleotide sequences recognised by the desired host, although such controls are generally available in the expression vector. For example, the nucleic acid molecule encoding a compound of the invention may be linked to or comprise a Kozak consensus ribosome binding sequence (such as GCCGCCACC) to

enhance translation.

The vector is then introduced into the host through standard techniques. Generally, not all of the hosts will be transformed by the vector. Therefore, it will be necessary to select for transformed host cells. One selection technique involves incorporating into the expression vector a nucleic acid sequence, with any necessary control elements, that codes for a selectable trait in the transformed cell, such as antibiotic resistance. Alternatively, the gene for such selectable trait can be on another vector, which is used to co-transform the desired host cell.

Host cells that have been transformed by the recombinant nucleic acid of the invention are then cultured for a sufficient time and under appropriate conditions known to those skilled in the art in view of the teachings disclosed herein to permit the expression of the polypeptide, which can then be recovered.

Many expression systems are known, including bacteria (for example *E. coli* and *Bacillus subtilis*), yeasts (for example *Saccharomyces cerevisiae* and *Pichia pastoris*), filamentous fungi (for example *Aspergillus*), plant cells, animal cells (for example COS-1, COS-7, CHO, NIH 3T3, NS0 and BHK cells) and insect cells (for example Drosophila, SF9 cells).

Those vectors that include a replicon such as a procaryotic replicon can also include an appropriate promoter such as a procaryotic promoter capable of directing the expression (transcription and translation) of the genes in a bacterial host cell, such as *E. coli*, transformed therewith.

A promoter is an expression control element formed by a DNA sequence that permits binding of RNA polymerase and transcription to occur. Promoter sequences compatible with exemplary bacterial hosts are typically provided in plasmid vectors containing convenient restriction sites for insertion of a DNA segment of the present invention.

Typical procaryotic vector plasmids are pUC18, pUC19, pBR322 and pBR329 (available from Biorad Laboratories, Richmond, CA, USA), p*Trc*99A and pKK223-3 (available from Pharmacia Piscataway, NJ, USA) and the pET system (T7 promoter, Novagen Ltd).

A typical mammalian cell vector plasmid is pSVL available from Pharmacia, Piscataway, NJ, USA. This vector uses the SV40 late promoter to drive expression of cloned genes, the highest level of expression being found in T antigen-producing cells, such as COS-1 cells.

An example of an inducible mammalian expression vector is pMSG, also available from Pharmacia. This vector uses the glucocorticoid-inducible promoter of the mouse mammary tumour virus long terminal repeat to drive expression of the cloned gene.

Useful yeast plasmid vectors are pRS403-406 and pRS413-416 and are generally available from Stratagene Cloning Systems, La Jolla, CA 92037, USA. Plasmids pRS403, pRS404, pRS405 and pRS406 are Yeast Integrating plasmids (YIps) and incorporate the yeast selectable markers *his3, trp1, leu2* and *ura3*. Plasmids pRS413-416 are Yeast Centromere plasmids (YCps).

Further useful vectors for transformation of yeast cells, such as *Pichia*, include the 2μ plasmid pYX243 (available from R and D Systems Limited) and the integrating vector pPICZ series (available from Invitrogen).

A variety of methods have been developed to operatively link DNA to vectors via complementary cohesive termini. For instance, complementary homopolymer tracts can be added to the DNA segment to be inserted to the vector DNA. The vector and DNA segment are then joined by hydrogen bonding between the complementary homopolymeric tails to form recombinant DNA molecules.

Synthetic linkers containing one or more restriction sites provide an alternative method of joining the DNA segment to vectors. The DNA segment, generated by endonuclease restriction digestion as described earlier, is treated with bacteriophage T4 DNA polymerase or *E. coli* DNA polymerase I, enzymes that remove protruding, 3'-single-stranded termini with their 3'-5'-exonucleolytic activities, and fill in recessed 3'-ends with their polymerizing activities.

The combination of these activities therefore generates blunt-ended DNA segments. The blunt-ended segments are then incubated with a large molar excess of linker molecules in the presence of an enzyme that is able to catalyze the ligation of blunt-ended DNA molecules, such as bacteriophage T4 DNA ligase. Thus, the products of the reaction are DNA segments carrying polymeric linker sequences at their ends. These DNA segments are then cleaved with the appropriate restriction enzyme and ligated to an expression vector that has been cleaved with an enzyme that produces termini compatible with those of the DNA segment.

Synthetic linkers containing a variety of restriction endonuclease sites are commercially available from a number of sources including International Biotechnologies Inc, New Haven, CN, USA.

A desirable way to modify the nucleic acid encoding the compound of the invention or a portion thereof is to use the polymerase chain reaction as disclosed by Saiki *et al* (1988) *Science* **239**, 487-491.

In this method the nucleic acid to be enzymatically amplified is flanked by two specific oligonucleotide primers which themselves become incorporated into the amplified nucleic acid. The said specific primers may contain restriction endonuclease recognition sites which can be used for cloning into expression vectors using methods known in the art.

Exemplary genera of yeast contemplated to be useful in the practice of the present invention are *Pichia, Saccharomyces, Kluyveromyces, Candida, Torulopsis, Hansenula, Schizosaccharomyces, Citeromyces, Pachysolen, Debaromyces, Metschunikowia, Rhodosporidium, Leucosporidium, Botryoascus, Sporidiobolus, Endomycopsis*, and the like. Preferred genera are those selected from the group consisting of *Pichia, Saccharomyces, Kluyveromyces, Yarrowia* and *Hansenula*. Examples of *Saccharomyces* are *Saccharomyces cerevisiae*, *Saccharomyces italicus* and *Saccharomyces rouxii*. Examples of *Kluyveromyces* are *Kluyveromyces fragilis* and *Kluyveromyces lactis*. Examples of *Hansenula* are *Hansenula polymorpha, Hansenula anomala* and *Hansenula capsulata*. *Yarrowia lipolytica* is an example of a suitable *Yarrowia* species.

Methods for the transformation of *S. cerevisiae* are taught generally in EP 251 744, EP 258 067 and WO 90/01063, all of which are incorporated herein by reference.

Suitable promoters for *S. cerevisiae* include those associated with the *PGK1* gene, *GAL1* or *GAL10* genes, *CYC1*, *PHO5*, *TRP1*, *ADH1*, *ADH2*, the genes for glyceraldehyde-3-phosphate dehydrogenase, hexokinase, pyruvate decarboxylase, phosphofructokinase, triose phosphate isomerase, phosphoglucose isomerase, glucokinase, α-mating factor pheromone, a-mating factor pheromone, the *PRB1* promoter, the *GUT2* promoter, and hybrid promoters involving hybrids of parts of 5' regulatory regions with parts of 5' regulatory regions of other promoters or with upstream activation sites (e.g. the promoter of EP-A-258 067).

The transcription termination signal is preferably the 3' flanking sequence of a eukaryotic gene which contains proper signals for transcription termination and polyadenylation. Suitable 3' flanking sequences may, for example, be those of the gene naturally linked to the expression control sequence used, i.e. may correspond to the promoter. Alternatively, they may be different in which case the termination signal of the *S. cerevisiae* *AHD1* gene is preferred.

The present invention also relates to a host cell transformed with a polynucleotide vector construct of the present invention. The host cell can be either procaryotic or eukaryotic. Bacterial cells are preferred procaryotic host cells and typically are a strain of *E. coli* such as, for example, the *E. coli* strains DH5 available from Bethesda Research Laboratories Inc., Bethesda, MD, USA, and RR1 available from the

American Type Culture Collection (ATCC) of Rockville, MD, USA (No ATCC 31343). Preferred eukaryotic host cells include yeast and mammalian cells, preferably vertebrate cells such as those from a mouse, rat, monkey or human fibroblastic cell line. Preferred eukaryotic host cells include Chinese hamster ovary (CHO) cells available from the ATCC as CCL61, NIH Swiss mouse embryo cells NIH/3T3 available from the ATCC as CRL 1658 and monkey kidney-derived COS-1 cells available from the ATCC as CRL 1650 or WSØ cells.

Transformation of appropriate cell hosts with a nucleic acid constructs of the present invention is accomplished by well known methods that typically depend on the type of vector used. With regard to transformation of procaryotic host cells, see, for example, Cohen *et al*, *Proc. Natl. Acad. Sci. USA*, **69**: 2110 (1972); and Sambrook *et al*, *Molecular Cloning, A Laboratory Manual*, Cold Spring Harbor Laboratory, Cold Spring Harbor, NY (1989). Transformation of yeast cells is described in Sherman *et al*, *Methods In Yeast Genetics, A Laboratory Manual*, Cold Spring Harbor, NY (1986). The method of Beggs, *Nature*, **275**: 104-109 (1978) is also useful. With regard to vertebrate cells, reagents useful in transfecting such cells, for example calcium phosphate and DEAE-dextran or liposome formulations, are available from Stratagene Cloning Systems, or Life Technologies Inc, Gaithersburg, MD 20877, USA.

Successfully transformed cells, *i.e.* cells that contain a nucleic acid construct of the present invention, can be identified by well known techniques. For example, cells resulting from the introduction of an expression construct of the present invention can be grown to produce the

polypeptide of the invention. Cells can be harvested and lysed and their DNA content examined for the presence of the DNA using a method such as that described by Southern, *J. Mol. Biol.*, **98**: 503 (1975) or Berent *et al*, *Biotech.*, **3**: 208 (1985). Alternatively, the presence of the protein in the supernatant can be detected using antibodies as described below.

In addition to directly assaying for the presence of recombinant nucleic acid, successful transformation can be confirmed by well known immunological methods when the recombinant nucleic acid is capable of directing the expression of the protein. For example, cells successfully transformed with an expression vector produce proteins displaying appropriate antigenicity. Samples of cells suspected of being transformed are harvested and assayed for the protein using suitable antibodies.

Thus, in addition to the transformed host cells themselves, the present invention also contemplates a culture of those cells, preferably a monoclonal (clonally homogeneous) culture, or a culture derived from a monoclonal culture, in a nutrient medium. Preferably, the culture also contains the protein.

Nutrient media useful for culturing transformed host cells are well known in the art and can be obtained from several commercial sources.

A third aspect of the invention provides a vector comprising a nucleic acid according to the second aspect of the invention.

A fourth aspect of the invention provides a host cell comprising a vector according to the third aspect of the invention.

Preferably, the host cell is a mammalian cell.

More preferably the host cell is NS0 or CHO.

A fifth aspect of the invention provides a pharmaceutical composition comprising a compound according to the first aspect of the invention and a pharmaceutically acceptable carrier.

The compounds and compositions of the invention are administered in any suitable way, usually parenterally, for example intravenously, intraperitoneally or, preferably (for bladder cancer), intravesically (*i.e.* into the bladder), in standard sterile, non-pyrogenic formulations of diluents and carriers, for example isotonic saline (when administered intravenously).

A sixth aspect of the invention provides a compound according to the first aspect of the invention for use in medicine.

The compounds and compositions of the invention may be used to treat a patient with any disease involving a dysfunction of a population of cells expressing PEM, said compounds and compositions selectively targeting and destroying said population of cells within a patient. For example, said compounds and compositions may be used in the treatment of cancer, *e.g.* cancer of the breast, ovaries, lung, stomach, intestines, blood *etc.* Thus, anti-tumour cell antigen antibodies can be used to deliver a cytotoxic portion with endonuclease activity to a tumour cell. Antibodies that are internalised upon contact with the target antigen are used, such that the

cytotoxic portion enters the cytosol of the tumour cell, where it can trigger cell death.

In principle, the compounds and compositions of the invention may be used to treat any mammal, including pets such as dogs and cats and agriculturally important animals such as cows, horses, sheep and pigs.

Preferably, the patient is human.

A seventh aspect of the invention provides the use of a compound according to first aspect of the invention in the preparation of a medicament for treating a mammal having said target cells to be destroyed.

Preferably, the medicament is for treating cancer, such as ovarian cancer.

A eighth aspect of the invention provides a method of treating a mammal having target cells to be destroyed, the method comprising administering a compound according to the first aspect of the invention to said mammal.

In a preferred embodiment of the seventh and eighth aspects of the invention, the mammal is a human.

Preferably, the target cells to be destroyed are cancer cells. More preferably, the cancer cells are epithelial cancer cells, such as ovarian, gastric, colorectal and/or pancreatic cancer cells. Most preferably, the cancer cells are ovarian cancer cells.

The invention will now be described in detail with reference to the following figures and examples:

Figure 1 shows the complete coding sequence of human DNAse I.

Figure 2 shows (A) the mature DNAse peptide I sequence used in the exemplary Ab-DNase and Fab-DNase constructs, and (B) a truncated DNAse peptide I sequence encoded by a nucleotide sequence comprising a Kozak sequence (underlined).

Figure 3 shows (A) the nucleotide sequence encoding the humanised HMFG1 light chain including leader peptide, (B) the nucleotide sequence of (A) further comprising a Kozak sequence (underlined), (C) the amino acid sequence of the humanised HMFG1 light chain including leader peptide (shaded) and (D) the nucleotide sequence encoding the humanised HMFG1 heavy chain including leader peptide,

Figure 4 shows the linker and hinge-linker oligonucleotides used in (A) the whole antibody-DNase and (B) the Fd-DNase exemplary constructs. Note, in Figure 4(A) a deletion of one or more codons between the HMFG1 hinge and the linker is represented as $\triangle$G.

Figure 5 shows nucleotide sequences (A and B) encoding a humanised HMFG-1 Fd/DNase I fusion pAS23 comprising a leader sequence (underlined) and a linker sequence (double-underlined). Figure 5(C) shows the nucleotide sequence of (B) further comprising a Kozak sequence (underlined). Figure (D) shows the amino acid sequence of a humanised HMFG-1 Fd/DNase I fusion.

Figure 6 shows (A), (B) and (C) shows the nucleotide sequences of Figure 5 (A), (B) and (C), respectively, further comprising an SV40 NLS (double underlined) (pAS27). Figure (D) shows the amino acid sequence of a humanised HMFG-1 Fd/DNase I fusion comprising an SV40 NLS (double underlined).

Figure 7 shows (A) the nucleotide sequence and (B) the translated amino acid sequence of an exemplary HMFG-1 heavy chain/DNase I fusion pAS34 (as used in 'Ab-DNase' in Example 2), comprising a leader sequence (underlined) and a linker sequence (double-underlined).

Figure 8 shows (A) the nucleotide sequence and (B) the translated amino acid sequence of an exemplary HMFG-1 heavy chain/DNase I fusion pAS35, comprising a leader sequence (underlined) and a linker sequence (double-underlined). The lower case 'g' represents a silent mutation caused by PCR amplification.

Figure 9 shows (A) the nucleotide sequence and (B) the translated amino acid sequence of an exemplary HMFG-1 heavy chain/DNase I fusion pAS36, comprising a leader sequence (underlined) and a linker sequence (double-underlined). The lower case 'c' represents a silent mutation caused by PCR amplification.

Figure 10 shows (A) the nucleotide sequence and (B) the translated amino acid sequence of an exemplary HMFG-1 heavy chain/DNase I fusion pAS37, comprising a leader sequence (underlined), a linker sequence (double-underlined) and an NLS sequence (triple underlined).

Figure 11 shows (A) the nucleotide sequence and (B) the translated amino acid sequence of an exemplary HMFG-1 heavy chain/DNase I fusion pAS38, comprising a leader sequence (underlined), a linker sequence (double-underlined) and an NLS sequence (triple underlined). The lower case 'g' represents a silent mutation caused by PCR amplification.

Figure 12 shows (A) the nucleotide sequence and (B) the translated amino acid sequence of an exemplary HMFG-1 heavy chain/DNase I fusion pAS39, comprising a leader sequence (underlined), a linker sequence (double-underlined) and an NLS sequence (triple underlined). The lower case 'c' represents a silent mutation caused by PCR amplification.

Figure 13 shows nucleotide sequences (A and B) encoding a humanised HMFG-1 Fd/DNase I fusion pAS101 comprising a short leader sequence (underlined) and a linker sequence (double-underlined). Figure 13(C) shows the nucleotide sequence of (B) further comprising a Kozak sequence (underlined). Figure (D) shows the amino acid sequence of a humanised HMFG-1 Fd/DNase I fusion.

Figure 14 shows nucleotide sequences (A and B) encoding a humanised HMFG-1 Fd/DNase I fusion pAS102 comprising a leader sequence (underlined) and a hybrid hinge + linker sequence (double-underlined). Figure 14(C) shows the nucleotide sequence of (B) further comprising a Kozak sequence (underlined) (construct designated pAS302 in Example 2). Figure (D) shows the amino acid sequence of a humanised HMFG-1 Fd/DNase I fusion.

Figure 15 shows nucleotide sequences (A and B) encoding a humanised HMFG-1 Fd/DNase I fusion pAS103 comprising a leader sequence (underlined) and a hybrid hinge + short linker sequence (double-underlined). Figure 15(C) shows the nucleotide sequence of (B) further comprising a Kozak sequence (underlined). Figure (D) shows the amino acid sequence of a humanised HMFG-1 Fd/DNase I fusion.

Figure 16 shows nucleotide sequences (A and B) encoding a humanised HMFG-1 Fd/DNase I fusion pAS104 comprising a leader sequence (underlined) and a hybrid hinge + mutated short linker sequence (double-underlined). Figure (C) shows the amino acid sequence of a humanised HMFG-1 Fd/DNase I fusion. Mutations (compared to pAS103) at positions 775 and 924 are shaded.

Figure 17 shows nucleotide sequences (A and B) encoding a humanised HMFG-1 Fd/DNase I fusion pAS105 comprising a leader sequence (underlined), a short linker sequence (double-underlined) and an NLS sequence (triple underlined). Figure 17(C) shows the nucleotide sequence of (B) further comprising a Kozak sequence (underlined). Figure (D) shows the amino acid sequence of a humanised HMFG-1 Fd/DNase I fusion.

Figure 18 shows nucleotide sequences (A and B) encoding a humanised HMFG-1 Fd/DNase I fusion pAS106 comprising a leader sequence (underlined), a hybrid hinge + linker sequence (double-underlined) and an NLS sequence (triple underlined). Figure 18(C) shows the nucleotide sequence of (B) further comprising a Kozak sequence (underlined). Figure (D) shows the amino acid sequence of a humanised HMFG-1

Fd/DNase I fusion.

Figure 19 shows nucleotide sequences (A and B) encoding a humanised HMFG-1 Fd/DNase I fusion pAS107 comprising a leader sequence (underlined), a hybrid hinge + short linker sequence (double-underlined) and an NLS sequence (triple underlined). Figure 19(C) shows the nucleotide sequence of (B) further comprising a Kozak sequence (underlined). Figure (D) shows the amino acid sequence of a humanised HMFG-1 Fd/DNase I fusion.

Figure 20 shows a schematic diagram of the pEE6 expression vector used in the exemplary constructs.

Figure 21 shows autoradiographs from immuno-precipitation experiments with metabolically labelled transient transfectants:

GEL A

Lane 1 shows the precipitation of supernatant from mock-transfected cells.

Lane 2 is from cells transfected with hHMFG-1 (construct 6) giving expected molecular weights of about 51.2 and 26.4 kDa for the heavy and light chains, respectively.

Lane 3 shows construct 34 antibody construct which has human DNase I fused to the C-terminus of the heavy chain gene. As expected, the size of the heavy chain gene has increased to about 80.7 kDa.

Samples from whole antibody DNase I constructs 35, 36 and 39 were run on the gel (Lanes 4 to 6) but were not sufficiently well

expressed to be visible, in this experiment.

In subsequent experiments using this method, construct 39 was detectable but weak, and constructs 35 and 36 were detectable but very weak. Constructs 37 and 38 have not been tested in this assay system.

Lanes 8 to 10 are fusion of humanised HMFG1 $F(ab')_2$ with human DNase I (constructs 41, 23 and 102, respectively). $F(ab')_2$ alone was included in this set of experiments (lane 7, construct 41) but did not express, this was included in later experiments (see gels C and D).

In addition to the light chain (about 26.4 kDa) and the Fd-DNAse I fusion (about 56.6 kDa), a third major band is observed at around 40 kDa. Interestingly, this band is observed in the humanised HMFG-1 fusions but not in the antibody alone. Since an anti-$F(ab')_2$ antibody was used for immuno-precipitation, it is unlikely that this can be proteolysis between immunoglobulin and DNase I sequence. It probably represents a population of polypeptide produced by premature transcriptional termination (due to DNase I sequence in the 3'-end of the fusion mRNA).

GEL B

This is the non-reducing gel counterpart to gel A, described above. Lane 1 is the mock-transfected control cells and lanes 2 and 3 are from the cells transfected with humanised HMFG1 alone (construct 6) and the humanised HMFG-1 fused at the C-terminus to human DNase I, respectively. As before, lanes 4 to 6 are from cell supernatants from cells transfected with constructs 35, 36 and 39. The gel shows that both the whole antibody and the antibody-DNase I fusion are assembled, with the DNase fusion giving a higher

molecular weight compared to the antibody alone.

Figure 22 shows a typical standard curve used to determine the concentration of PDTRP-binding material in the supernatants of transiently transfected L761h cells. Each point on the curve has been determined twice.

Figure 23 shows typical standard curves used to determine the concentration of bovine DNAse I.

Figure 24 shows corrected DNase I activity in transiently expressed humanised HMFG1 whole antibody-human DNAse I fusions (*i.e.* pAS34, pAS34, pAS35 and pAS6[control]).

Figure 25 shows the corrected DNAse I activity in transiently expressed humanised HMFG1 $F(ab')_2$-human DNase I fusions (*i.e.* pAS101, pAS102, pAS103 and pAS41[control]).

Figure 26 shows results of the cytotoxicity assay.

Figure 27 shows the % of MCF7 cells killed after incubation with the exemplary constructs.

Figure 28 shows a schematic diagram of (A) Ab-DNase and (B) Fab-DNase.

Figure 29 shows a schematic diagram of vector pAS34K encoding Ab-DNase (*i.e.* pAS34 as shown in Figure 7b plus Kozak sequence).

Figure 30 shows a schematic diagram of vector pAS302 encoding Fab-DNase.

Figure 31 shows (A) the elution profile from Protein-L column and (B) size exclusion chromatogram for Fab-DNase.

Figure 32 shows (A) the elution profile from Protein-A column and (B) size exclusion chromatogram for Ab-DNase.

Figure 33 shows the SDS-PAGE stained gels for (A) Ab-DNase and (B) Fab-DNase.

Figure 34 shows (A) standard curve for bovine DNase concentration AND (B) DNase activity measurements at 3 hours and 6 hours.

Figure 35 shows (A) PEM expression on OVCAR 3 and A375 cells, as measured by ELISA using hHMFG-1 and AD-DNase antibodies, and (B) cytotoxicity measurements.

EXAMPLES

Example 1

(A) Mammalian expression of humanised HMFG1-DNase constructs

The human HMFG1 light and heavy chain (with or without engineering a fusion to human DNase I), were cloned into the pEE6 expression vector system for expression in mammalian CHO or myeloid NS0 cells (see figure 20). The vector system was originally developed by Celltech Ltd (UK) and is now owned by al-Lonza (see Young & Owens, 1994, *J. Immunol. Meth.* **168**:149-165). The vector consists of two human cytomegalovirus promoters (hCMV) for both the heavy and light chain genes. Each transcription unit is completed by the poly-adenylation signal (pA) with an optional immunoglobulin terminator sequence (Ig term.) located between the heavy and light chain transcription units. Propagation in *E.coli* can be selected for by the presence on an ampicillin resistance gene (not shown in Fig 20). The inclusion of a glutamine synthetase gene (GS) in the vector allows the stable NS0 transfectomas to be selected by growth in glutamine free media, since NS0 cells are GS^- and cannot otherwise grow in glutamine free media.

Exemplary humanized HMFG1-DNAse I fusion constructs of the invention are detailed in figures 5 to 19.

(B) Immuno-precipitation of metabolically labelled transient transfectants

CHO-L761h cells (Cockett *et al.*, 1990, *Nuc. Acids Res.* **19**:319-325)

were transfected, according to the modification of Gorman et al, 1985), with expression vectors containing either whole HMFG1 antibody or $F(ab')_2$ fragment of the antibody along with the various fusion constructs of their respective heavy chains and human DNase I. The cells were then incubated with either 50 μCi ^{35}S methione for 72 h in methionine-free medium. Secreted product was precipitated with a rabbit anti-human $F(ab')_2$ antibody bound to protein A Sepharose. Bound material was eluted in either reducing or non-reducing SDS-PAGE loading buffer and run on gels. The autoradiographs (see Figure 21) above were generated from those gels after drying them.

(C) Estimation of the efficiency of DNase constructs in supernatants

Introduction

This set of experiments was designed to standardise the amount of construct in a given DNase I activity assay and to allow us to comment on the amount of activity a particular construct possesses. Given that the antibody-DNase I fusions are so different to the $F(ab')_2$-DNase I fusions it is best not to compare the two groups. Once we have purified the protein, we will have a better idea of the exact molecular configuration of all species. Then, and only then, will it be sensible to compare amongst groups.

Determination of concentration of constructs

The concentration of constructs in supernatants from transiently transfected L761H cells was determined in a PDTRP-binding ELISA. To

each well of a Maxisorb 96-well ELISA plate (Nunc) was added 100 μl of carbonate buffer containing 100 ng of recombinant GST-$(PDTRP)_7$ fusion protein (Gendler *et al.*, 1990, *J. Mol. Biol.* **265**:15286-93). After overnight binding at 4°C, the plate was washed three times in PBS-Tween (*i.e.* PBS containing 0.05% Tween-20). The plate was then blocked with three 3-minute washes of PBS-Tween containing 1% BSA.

For each construct, 100 μl of supernatant was added to a well on the plate. In addition, hHMFG-1 of known concentration was serially diluted down the plate using doubling dilutions in 100 μl of PBS-Tween per well. The plate was incubated for a further 1 h at 30°C, then 200 ng of MC135 anti-human kappa light chain antibody (binding site) in 100 μl of PBS-Tween was added to each well for 1 h at 30°C. After three 3-minute washes in PBS-Tween, 100 μl of anti-mouse IgG-peroxidase conjugate (Jackson 315-035-045), diluted 1:2000 in PBS-Tween, was added to each well and incubated for 1 h at 30°C. Following a final set of three 3-minute washes in PBS-Tween, 100 μl of TMB substrate (Sigma) was added to each well of the plate and, after a colour developed, the optical density at 630 nm of the solution in each well of the plate was determined.

Results

(see Figure 22)

(D) Corrected bovine DNase I standard curves and DNase assay

DNase activity was determined using a modification of the methyl green-DNA complex degradation method (Sinicropi *et al.*, 1994, *Analyt.*

Biochem. **222**:351-358). Briefly, a 1:1 solution of the assay buffer and methyl green-salmon sperm DNA complex was mixed together to give a total volume of 0.2 ml. To this, 0.1 ml of tissue culture supernatant from transiently transfected CHO-L761h cells was added and the mixture incubated at 37°C. DNA cleavage by DNase results in a reduction in absorbance at 620 nm. Figure 23 shows a standard curve produced with various concentrations of bovine DNase I over a number a time point.

Figures 24 and 25 show DNAse activity for the whole HMFG1 antibody- and $F(ab')_2$ - DNase fusions, respectively.

(E) Cytotoxicity of DNAse constructs

Method

DNase constructs were transfected into CHO L761h cells using a calcium phosphate co-precipitation method (Gorman *et al.*, 1985, In: *DNA cloning* (2nd edition), Glover A(ed.), Academic Press, NY, 163-188). Included in the experiment were negative controls, consisting of cells transfected with TE buffer alone or with TE buffer and pEE6 expression vector. In addition to these controls, vectors that express hHMFG-1 (pAS6) and $F(ab')_2$ of hHMFG1 (both with specificity for PEM but without DNase I) were included.

The supernatant from these cells was harvested after 72 h of expression, followed by centrifugation to remove dead cells. MCF-7 cells were incubated for 1 h at 37°C with an aliquot of each of these supernatants. The amount of cellular lactate dehydrogenase (LDH) released from the

MCF-7 cells due to the cytotoxicity of the supernatant was determined using the CytoTox96 cytotoxic assay kit (Promega). Total lysis ('total LDH') was determined by measuring the target cell maximum LDH release using the kits lysis solution. The percentage of cells killed was then calculated as the proportion of the LDH released to the total LDH released. For each construct, the cytotoxicity assay was performed in quadruplicate, except for assay of pAS38 and 39, which were performed in triplicate. The values of LDH release for each construct were compared against either $F(ab')_2$ or whole antibody, or each other, using a one-tailed t-test in Excel.

Results

Figures 26 and 27 shows that there is negligible cell killing with either pAS6 (HMFG1 alone) or with pAS41 ($F(ab')_2$ alone). All of the hHMFG1 $F(ab')_2$-DNase I constructs kill significantly more cells than the $F(ab')_2$ fragment alone ($p < 0.00193$) and all of the antibody-DNase I constructs kill significantly more cells than antibody alone ($p < 0.00783$), except for perhaps pAS34 ($p < 0.021$).

(F) Use of the DNase-I/huHMFG-1 Fab fusion protein in the treatment of ovarian cancer

Patients diagnosed with ovarian cancer are treated by intravenous injection of the DNaseI/huHMFG-1 Fab fusion protein. Typically, a dose of between 1 to 100 mg will be administered weekly.

Therapeutic response is measured by the normal clinical procedures that

are well known in the art, for example radio-imaging methods.

Example 2

(A) Mammalian expression of humanised HMFG-1 / DNase constructs

In a second series of experiments, two further humanised HMFG-1/Dnase constructs were expressed in mammalian cells. The first construct encoded a fusion protein a complete hHMFG-1 antibody fused with human DNase, designated 'Ad-DNase'. The second construct encoded a fusion protein a Fab fragment of the hHMFG-1 antibody fused with human DNase, designated 'Fab-DNase'. Ad-Dnase and Fab-DNase are shown schematically in Figure 28.

Ad-DNase comprises an HMFG-1 light chain as shown in Figure 3(c) and an HMFG-1 heavy chain/DNase fusion as shown in Figure 7(b).

Fab-DNase comprises an HMFG-1 light chain as shown in Figure 3(c) and an HMFG-1 Fd chain/DNase fusion as shown in Figure 14(d).

The human HMFG1 heavy and light chain constructs were cloned into the pEE6 expression vector system for expression in mammalian CHO or myeloid NS0 cells, as described in Section (A) of Example 1. This vector consists of two human cytomegalovirus promoters (hCMV) for both the heavy and light chain genes. Each transcription unit is completed by the poly-adenylation signal (pA) with an optional immunoglobulin terminator sequence (Ig term.) located between the heavy and light chain transcription units. The vectors also comprise a 5'-UT Kozak sequence

(to enhance translation of the mRNA) and an ATG initiator codon upstream of both heavy and light chains.

The vectors encoding Ad-Dnase and Fab-DNase, designated pAS34K and pAS 302 respectively, are shown schematically in Figure 32.

Propagation in *E.coli* can be selected for by the presence on an ampicillin resistance gene. The inclusion of a glutamine synthetase gene (GS) in the vector allows the stable NS0 transfectomas to be selected by growth in glutamine free media, since NS0 cells are GS^- and cannot otherwise grow in glutamine free media.

These plasmids were co-transfected with a vector containing a neomycin resistance gene into CHO cells. Stable cell lines were generated for each of the constructs.

Clones were selected that expressed DNase activity and antigen (PEM)-binding activity.

(B) Purification of hHMFG-1/DNase constructs

The cells were routinely grown in:

DMEM (Gibco 10938-025)	500 ml
Non essential amino acids (Sigma M7145)	5 ml
Sodium pyruvate (Sigma S8636)	5 ml
Glutamine (G7513)	5 ml
Heat inactivated foetal calf serum	50 ml

Incubation was carried out at 37°C in 5% CO_2.

For production of the Ab-DNase fusion protein, W70 cells (CHO cells transfected with pAS34K) were maintained in flats and grown to confluency in T175 flasks. Each T175 flask was split between two 850 cm^2 roller bottles containing 100 ml of the aforementioned growth media. Each roller bottle was gassed with an 95% air 5% CO_2 mix for 1 minute and then sealed. They were rolled at a rate of 0.5 rpm and were gassed every other day as described earlier until the cultures were confluent. At this stage the medium was removed and 200 ml of harvest medium was replaced on the culture. This was the same medium but contained 2 mM sodium butyrate (with or without 10% heat inactivated FCS). The cells were then grown for a further 3-4 days before they were harvested. The medium was collected from the cells and dead cells were removed from the medium by centrifugation at 5000 rpm for 30 mins at 4°C. The spun medium (supernatant) was then filtered through a 0.2 micron filter unit, prior to applying to the affinity chromatography column.

The Fab-DNase fusion product was then purified by affinity chromatography using a Protein-L column (Protein L agarose, P3351 from Sigma Co, Poole, Dorset, UK), as follows:

1. Wash 1 ml of settled protein L agarose (P3351) with at least 5 volumes of phosphate buffered saline (PBS: 10 mM phosphate buffered saline, pH 7.4).
2. Dilute 1 ml supernatant with 9 ml PBS.

3. Mix diluted supernatant with protein-L agarose and incubate with gentle end over end mixing for 1 hour at room temperature.
4. Pack the slurry in a column and drain.
5. Wash away unbound proteins with 10-15 column volumes of PBS.
6. Elute bound protein with5 ml elution buffer (0.1 M glycine, pH 2.0, or 0.2 M citrate buffer, pH2.8).
7. Neutralise eluted material with Tris-base to achieve pH 7.5.

Figure 31(a) shows the elution profile of the Fab-DNase from the Protein-L column when eluted with 0.1 M glycine, pH 2.0.

Following purification, Fab-DNase was analysed by analytical size-exclusion chromatography on a Superdex-200 column.

Figure 31(b) shows the size-exclusion chromatogram obtained for the Fab-DNase.

The Ab-DNase fusion product was purified by affinity chromatography using a Protein-A sepharose column, as follows:

1. 25 ml of protein A sepharose fast flow resin (Amersham Pharmacia Biotech) in an XK26 column (Amersham Pharmacia Biotech) was equilibrated in 0.1M glycine, pH8.8, 0.5M NaCl.
2. Approximately 2 litres of sterile-filtered supernatant from cell line W70 (CHO cell line making 34K) was passed the column overnight at a low flow rate (1-2 ml/min).
3. The column was then washed down to base-line and was re-equilibrated in 0.15M disodium hydrogen phosphate, pH9.0 and the

bound 34K was eluted by running a gradient between this buffer (A) and a low pH buffer (B) which consisted of 0.1M citric acid, pH2.0, supplemented to 2 mM calcium chloride and 2 mM magnesium sulphate. The gradient was run over 100 ml at a flow rate of 4 ml/min and a further 50 ml of buffer B was run over the column at the completion of the gradient, also at 4 ml/min.

4. During the 100 ml gradient and the last 50 ml of buffer A fractions were collected. The peak fractions were identified and pooled and dialysed against 4 litres of 25 mM Hepes, pH7.5, 0.2 M NaCl, 1mM calcium chloride and 1mM magnesium sulphate. Dialysis was performed overnight at 4C.
5. The dialysate was concentrated on Centricon spin concentrators to a final concentration of 6-13 mg/ml. The concentration was determined by dividing by its extinction coefficient of 1.558 (calculated from the known sequence).

Figure 32(a) shows the elution profile of the Ab-DNase from the Protein-L column when eluted with a gradient of 0.15 M Na_2HPO_4, pH 9.0 to 0.1 M citric acid, pH 2.0 containing 2mM each of $CaCl_2$ and $MgCl_2$.

Figure 32(b) shows the size-exclusion chromatogram obtained for the Ab-DNase.

(C) Determination of concentration of fusion proteins

Prior to measuring DNase activity of the purified fusion proteins (see Section (E) below), the concentration of the proteins was determined by ELISA, as follows (see also Section (C) of Example 1).

Materials

1. 96 Well ELISA plates (Nunc F96 Maxisorp Cat No. 442404).
2. Bovine serum albumin (Sigma A-9647).
3. Coating buffer (Na2CO3 1.59 g/l, NaHC03 2.93 g/l, NaN3 0.2 g/l, pH9.6.
4. GST-MUC1-7TR antigen (1.5 mg/ml).
5. Anti-human kappa light-chain antibody GD12 (0.2 mg/ml, Binding Site, MC135).
6. Peroxidase-conjugated rabbit anti-mouse IgG (Jackson, 315-035-045).
7. TMB- substrate buffer (Sigma P-4417).
8. Tween 20 (Sigma P7949).
9. Purified humanised HMFG1 (1.4 mg/ml).

Method

Note all washes in this protocol consist of 3 x 3 min washes in PBS buffer (note: all PBS buffer contained 0.05 % Tween) and the plate was incubated in a lunch box containing moist tissue paper.

1. Coat 100 ng of antigen/100 μl coating buffer/well overnight at 4°C.
2. Wash the plate and block each well with 100 μl of PBS containing 0.05 % Tween, and 1% BSA for 1 h at 30°C. Wash plate afterwards.
3. A standard curve of humanised HMFG1 should be prepared

down the plate using doubling dilutions. Make each dilution in 100 μl PBS buffer and for the highest concentration in the curve use 1000 ng of antibody.

4. Incubate the plate for 2 h at 30°C, wash, and add 100 μl PBS containing 200 ng of the anti-human Kappa light chain antibody to each well of the plate. Incubate for a further 1 h at 30°C and then wash the plate.

5. Add 100 μl PBS containing the rabbit anti-mouse IgG-peroxidase conjugate (diluted 1:2000) to each well of the plate and incubate for 30 min at 30°C. Wash the plate and add 100 μl TMB- substrate-buffer to each well of the plate and allow the reaction to proceed in the dark at room temperature. When the blue colour has developed, read the plate at a wavelength of 630 nm.

(D) SDS-PAGE

Following purification of Ab-DNase and Fab-DNase, the fusion proteins were analysed by SDS-PAGE under non-reducing and reducing conditions, as described in Section (B) of Example 1.

In brief, affinity-purified material was used. In the case of the Ab-DNase fusion protein, this was from a sample dialysed and concentrated (as described in the protein A protocol above). In the case of the Fab-DNase, this was unconcentrated protein directly eluted from the protein L affinity column. 15 ul of the Fab-DNase protein-L eluate was mixed with 5 ul of either reducing or non-reducing loading buffer whereas 2 ul of the Ab-DNase protein A eluate (dialysed and concentrated) was mixed with 5 ul

of either reducing or non-reducing buffer. Both samples were boiled for 5 minutes and were loaded onto the gel. The gels were stained with Coomassie Brilliant Blue stain. The cells were not labelled with 35S-methionine (as in Example 1).

The SDS-PAGE autoradiograph for Ab-DNase is shown in Figure 33(a). Under reducing conditions, Ab-DNase produces a band of about 80 kDa, which corresponds to the expected size of the heavy chain-DNase fusion product (see lane 3). A further band of about 50 kDa is also observed, which is approximately the same molecular weight as the hHMFG-1 heavy chain (see lane 4).

The SDS-PAGE autoradiograph for Fab-DNase is shown in Figure 33(b). Under reducing conditions, Fab-DNase produces a band of about 55-60 kDa, which corresponds to the expected size of Fab-DNase (see lane 3). Under non-reducing conditions, a band of about 80-85 kDa is observed, which is the approximate molecular weight of Fab-DNase rather than $F(ab')_2$-DNase (see lane 4). Thus, the Fab-DNase appears to exist as a dimer of the hHMFG-1 light chains and the hHMFG-1 heavy chain/human DNase fusion, not a tetrameric $F(ab')_2$-DNase.

(E) Measurement of DNase activity of hHMFG-1/DNase constructs

DNase activity of the two fusion proteins was determined as described in Section (D) of Example 1. In brief, 0.1 ml of the purified protein was added to a 1:1 solution of assay buffer and methyl green-salmon sperm DNA complex, and the mixture incubated at 37°C. A reduction in absorbance at 620 nm is indicative of DNA activity.

A standard curve produced using bovine DNase I is shown in Figure 34(a).

Figure 34(b) shows the DNase activity of the Fab-DNase and Ab-DNase fusion proteins 3 h and 6 h after being added to the DNA, compared to a positive control of bovine DNase and a negative control of Fab only. Clearly, the DNase activity of the Fab-DNase and Ab-DNase fusion proteins is comparable to that of the bovine DNase positive control.

(F) Cytotoxicity of DNase activity of hHMFG-1/DNase constructs

Cytotoxocity of the Fab-DNase and Ab-DNase fusion proteins was analysed using two tumour cell lines, the human malignant melanoma cell line A375 and the human ovarian adenocarcinoma cell line OVCAR 3.

An initial cell-based ELISA was performed using hHMFG-1 antibodies to determine the level of expression of PEM (the MUC1 gene product) on these cells.

Cell-based PEM ELISA assay protocol

Materials and methods

1. Phosphate buffered saline tablets (Sigma P-4417)
2. 50% glutaraldehyde solution (BDH UN2810 Prod. 2868240)
3. sodium azide (Sigma S-8032)
4. Nunclon 96 well tissue culture plate (Nunc D167008)

5. BSA (Sigma A-9647)
6. OVCAR-3 ovarian cancer cells, A375 melanoma cancer cells both from ATCC
7. TMB substrate buffer (Sigma P-4417)
8. Tween 20 (Sigma P7949)
9. Purified humanised HMFG1 (1 mg/ml from ICRF)
10. RPMI 1640 media (Gibco 21875-034)

Protocol

1. The OVCAR-3 and A375 cells were grown in RPMI containing 20% and 10% FCS respectively at 37°C in 5% CO2 in a 96 well tissue culture plate, seeded at 106 cells/ml with 0.1 ml/well.
2. Excess media was removed and the plate was fixed with 0.05% glutaraldehyde in water for 1 hour at room temperature.
3. Excess glutaraldehyde/water solution was removed and the plates were washed three times with PBS containing 0.05% Tween 20. The plate was stored at 4°C until required in PBS with 0.02% sodium azide).
4. To use the plate, the plate was then washed with three washes of PBS containing 0.05% Tween 20, and the wells were blocked with 0.1 ml 5% BSA in PBS containing 0.05% Tween 20. The wells were blocked for 1 hour at 30°C.
5. They washed three times as described before. Serial dilutions of hHMFG1 were plated out on the wells from a maximum concentration of 2 μg/ml downward. Dilutions of constructs were also similarly plated onto the fixed cells. All dilutions were prepared in PBS containing 0.05% Tween 20.

6. The proteins were incubated with the fixed cells for 1 hour at 30°C and were again washed three times as described above.
7. Anti-human IgG-Fc peroxidase conjugate antibody (Jackson 209-035-103) was diluted to 1:2000 in PBS containing 0.05% Tween 20. This was incubated at 30°C for 30 minutes.
8. Once again the cells were washed as described as before. Then 0.1 ml TMB substrate was put in each well and the colour was developed at room temperature and the absorbance at 655 nm was determined.

For comparison, an additional ELISA using Ab-DNase was performed with the OVCAR 3 cells.

Antigen-bound hHMFG-1 and Ab-DNase was detected by a peroxidase-conjugated anti-human Fc antibody.

The results of the ELISA are shown in Figure 35, indicating that the OVCAR 3 cell line expresses high levels of PEM (as measured by both hHMFG-1 and Ab-DNase) while the A375 cell line expresses low levels of PEM (and hence can be used as a negative control in cytotoxicity experiments).

Cytotoxicity was measured using an LDH release assay, as described in Section (E) of Example 1. In brief, 10^5 cells per well of the A375 and OVCAR 3 cell lines were plated in a 96-well plate and grown for 24 hours. Fifteen microlitres of the purified fusion proteins (containing 200 ng of Ab-DNase or 100 ng of Fab-DNase) were added to the cells and incubated for 48 hours at 37°C. A negative control group of each cell

type was treated with 200 ng of the hHMFG-1 antibody (*i.e.* not fused to DNase).

Following the incubation period, 50 μl of the supernatant was removed and incubated with 50 μl of tetrazolium-containing substrate buffer for 30 minutes at 22°C. The reaction was stopped with stop buffer (Promega) and the absorbance of the reaction mixture at 490 nm measured.

Both Fab-DNase and Ab-DNase fusions show cell killing of OVCAR 3 cells as compared to the negative control hHMFG-1 treated cells. In contrast, killing of A375 cells by DNase fusions is negligible, consistent with negligible binding of the fusions to these cells.

1/113

Human DNase I

```
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ACCESSION   M55983
VERSION     M55983.1  GI:181623
KEYWORDS    DNase I.
SOURCE      Human pancreus, cDNA to mRNA.
  ORGANISM  Homo sapiens
            Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Mammalia;
            Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1039)
  AUTHORS   Shak,S., Capon,D.J., Hellmiss,R., Marsters,S.A. and Baker,C.L.
  TITLE     Recombinant human DNase I reduces the viscosity of cystic fibrosis
            sputum
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 87 (23), 9188-9192 (1990)
  MEDLINE   91067672
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//
```

Fig. 1

2/113

Human DNase I construct

```
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site
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NID         g181623
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SOURCE      Human pancreus, cDNA to mRNA.
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
REFERENCE   1  (bases 1 to 1039)
  AUTHORS   Shak,S., Capon,D.J., Hellmiss,R., Marsters,S.A. and Baker,C.L.
  TITLE     Recombinant human DNase I reduces the viscosity of cystic fibrosis
            sputum
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 87 (23), 9188-9192 (1990)
  MEDLINE   91067672
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Fig. 2(A)

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ACCESSION   -
KEYWORDS    -
SOURCE      -
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//
```

Fig. 2(B)

4/113

pAS6 – light chain

```
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ACCESSION   ____________________________________________
KEYWORDS
SOURCE
  ORGANISM
REFERENCE   1  (BASES 1 TO 342)
  AUTHORS   VERHOEYEN ET AL
  TITLE     CONSTRUCTION OF RESHAPED HMFG1 ETC
  JOURNAL   IMMUNOL. (1993):78, 364-370
COMMENT     SCANNED IN FROM JOURNAL
FEATURES
  SITES

This is the sequence of the HMFG1 light chain gene with the
Vnp  leader sequence attached. Translate from
residue 1. Note residue 399 is T > A in all clones leading
to R133 silent mutation (T in Verhoeyen paper)

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        481 CTGAATAACT TCTATCCCAG AGAGGCCAAA GTACAGTGGA AGGTGGATAA CGCCCTCCAA
        541 TCGGGTAACT CCCAGGAGAG TGTCACAGAG CAGGACAGCA AGGACAGCAC CTACAGCCTC
        601 AGCAGCACCC TGACGCTGAG CAAAGCAGAC TACGAGAAAC ACAAAGTCTA CGCCTGCGAA
        661 GTCACCCATC AGGGCCTGAG CTCGCCCGTC ACAAAGAGCT TCAACAGGGG AGAGTGTTAG
        721 A
//
```

Fig. 3(A)

5/113

```
LOCUS       HHMFG1KLC_     730 BP SS-DNA              SYN       29-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
     frag            10..730
                     /note="1 to 721 of hHMFG1light chain"
     frag            10..730
                     /note="1 to 72 of 104linker"
     frag            join(10..>63,<65..81)
                     /note="1 to 72 of 103linker [Split]"
     frag            join(10..>60,<61..>63,<65..81)
                     /note="1 to 78 of 102linker [Split]"
BASE COUNT      198 A    208 C    184 G    140 T      0 OTHER
ORIGIN      -
        1 GCCGCCACCA TGGGATGGAG CTGTATCATC CTCTTCTTGG TAGCAACAGC TACAGGTGTC
       61 CACTCCGACA TCCAGATGAC CCAGAGCCCA AGCAGCCTGA GCGCCAGCGT GGGTGACAGA
      121 GTGACCATCA CCTGTAAGTC CAGTCAGAGC CTTTTATATA GTAGCAATCA AAAGATCTAC
      181 TTGGCCTGGT ACCAGCAGAA GCCAGGTAAG GCTCCAAAGC TGCTGATCTA CTGGGCATCC
      241 ACTAGGGAAT CTGGTGTGCC AAGCAGATTC AGCGGTAGCG GTAGCGGTAC CGACTTCACC
      301 TTCACCATCA GCAGCCTCCA GCCAGAGGAC ATCGCCACCT ACTACTGCCA GCAATATTAT
      361 AGATATCCTC GGACGTTCGG CCAAGGGACC AAGGTGGAAA TCAAACGAAC TGTGGCTGCA
      421 CCATCTGTCT TCATCTTCCC GCCATCTGAT GAGCAGTTGA AATCTGGAAC TGCCTCTGTT
      481 GTGTGCCTGC TGAATAACTT CTATCCCAGA GAGGCCAAAG TACAGTGGAA GGTGGATAAC
      541 GCCCTCCAAT CGGGTAACTC CCAGGAGAGT GTCACAGAGC AGGACAGCAA GGACAGCACC
      601 TACAGCCTCA GCAGCACCCT GACGCTGAGC AAAGCAGACT ACGAGAAACA CAAAGTCTAC
      661 GCCTGCGAAG TCACCCATCA GGGCCTGAGC TCGCCCGTCA CAAAGAGCTT CAACAGGGGA
      721 GAGTGTTAGA
//
```

Fig. 3(B)

6/113

HMFG-1 light chain with Vnp Leader (shaded)

MGWSCIILFLVATATGVHSDIQMTQSPSSLSASVGDRVTITCKSSQSL
LYSSNQKIYLAWYQQKPGKAPKLLIYWASTRESGVPSRFSGSGSGT
DFTFTISSLQPEDIATYYCQQYYRYPRTFGQGTKVEIKRTVAAPSVFI
FPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNALQSGNSQESV
TEQDSKDSTYSLSSTLTLSKADYEKHKVYACEVTHQGLSSPVTKSFN
RGEC

Fig. 3(C)

7/113

pAS6 – heavy chain

```
LOCUS       HHMFG1HC.D   1404 bp    DNA                     14-AUG-1998
DEFINITION   HUMANISED HMFG1 heavy chain
ACCESSION   HHMFG1H
KEYWORDS
SOURCE
  ORGANISM
REFERENCE
  AUTHORS   VERHOEYEN ET AL
  TITLE     CONSTRUCTION OF RESHAPED HMFG1 etc
  JOURNAL   IMMUNOL. (1993):78, 364-370
COMMENT     VH domain SCANNED IN FROM JOURNAL
FEATURES    AA RESIDUE 235 HAS NOT BEEN CHANGED TO KABAT (I.E. V TO A)
FEATURES    Residue 963 is G > T leading to silent mutation in all clones
  SITES     Note
BASE COUNT      333 a    439 c    379 g    253 t
ORIGIN           ?
```

LEADER

```
     1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
    61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
   121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
   181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
   241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
   301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
   361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
   421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
   481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
   541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
   601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
   661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
   721 GACAAAACTC ACACATGCCC ACCGTGCCCA GCACCTGAAC TCCTGGGGGG ACCGTCAGTC
   781 TTCCTCTTCC CCCCAAAACC CAAGGACACC CTCATGATCT CCCGGACCCC TGAGGTCACA
   841 TGCGTGGTGG TGGACGTGAG CCACGAAGAC CCTGAGGTCA AGTTCAACTG GTACGTGGAC
   901 GGCGTGGAGG TGCATAATGC CAAGACAAAG CCGCGGGAGG AGCAGTACAA CAGCACGTAC
   961 CGTGTGGTCA GCGTCCTCAC CGTCCTGCAC CAGGACTGGC TGAATGGCAA GGAGTACAAG
  1021 TGCAAGGTCT CCAACAAAGC CCTCCCAGCC CCCATCGAGA AAACCATCTC CAAAGCCAAA
  1081 GGGCAGCCCC GAGAACCACA GGTGTACACC CTGCCCCCAT CCCGGGATGA GCTGACCAAG
  1141 AACCAGGTCA GCCTGACCTG CCTGGTCAAA GGCTTCTATC CCAGCGACAT CGCCGTGGAG
  1201 TGGGAGAGCA ATGGGCAGCC GGAGAACAAC TACAAGACCA CGCCTCCCGT GCTGGACTCC
  1261 GACGGCTCCT TCTTCCTCTA CAGCAAGCTC ACCGTGGACA AGAGCAGGTG GCAGCAGGGG
  1321 AACGTCTTCT CATGCTCCGT GATGCATGAG GCTCTGCACA ACCACTACAC GCAGAAGAGC
  1381 CTCTCCCTGT CTCCGGGTAA ATGA
```

//

Antibody DNase Fusions Made Here
(eg pAS34----39.)

End of lower hinge region of heavy chain. PAPE Amino Acid Seq. Fab'$_2$ fusions were made at this point.

Those with HYBRID HINGES are altered further up i.e.

This part GACAAAACTGACACA
D K T H T

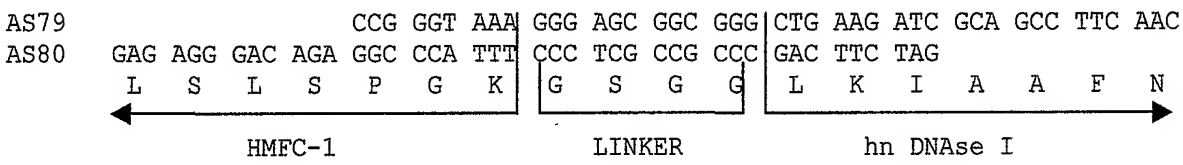
After this sequence you get the HYBRID HINGE + LINKER SEQUENCES
Then DNAse I (eg Fab-DNase construct pAS302)

Fig. 3(D)

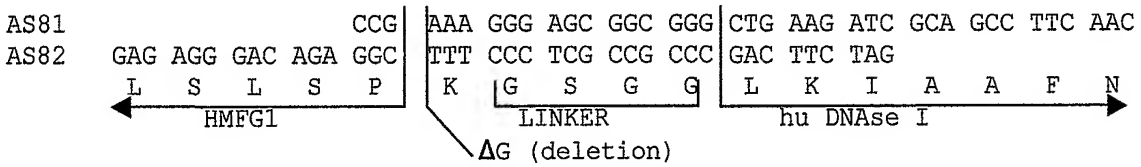
Fig. 4(A)

Oligos involved in the fusion of whole antibody-DNase

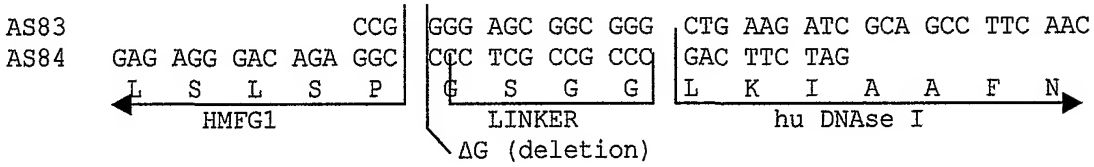
Constructs pAS34/37



Constructs pAS35/38

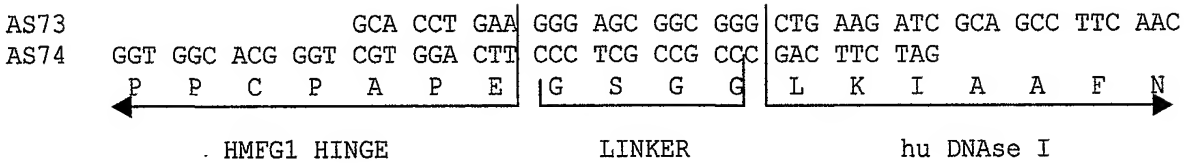


Constructs pAS36/39



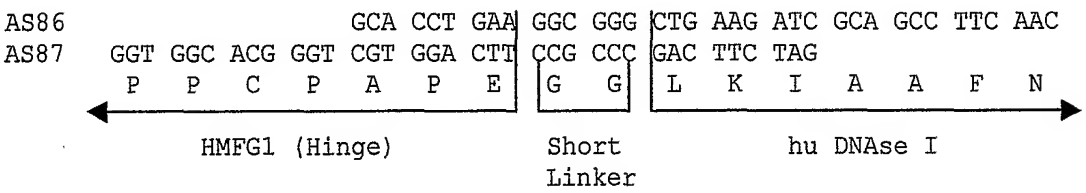
Oligos involved in the fusion of Fab'2-DNaseI

Constructs pAS23/27

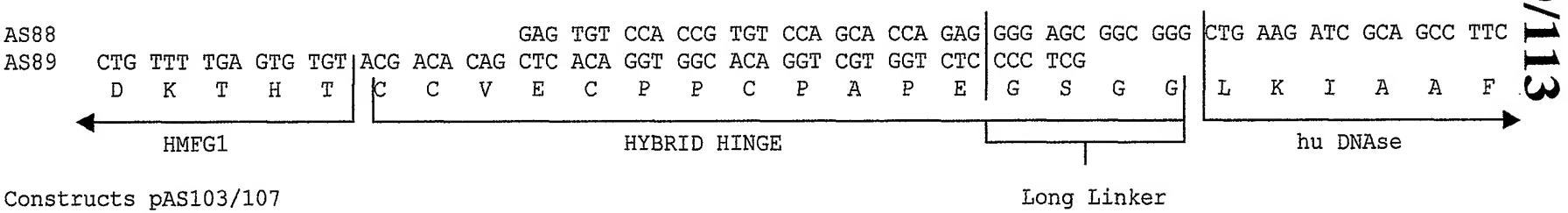


Oligos involved in the fusion of new Fab'2-DNaseI molecules (5.7.99)

Constructs pAS101/105



Constructs pAS102/106



Constructs pAS103/107

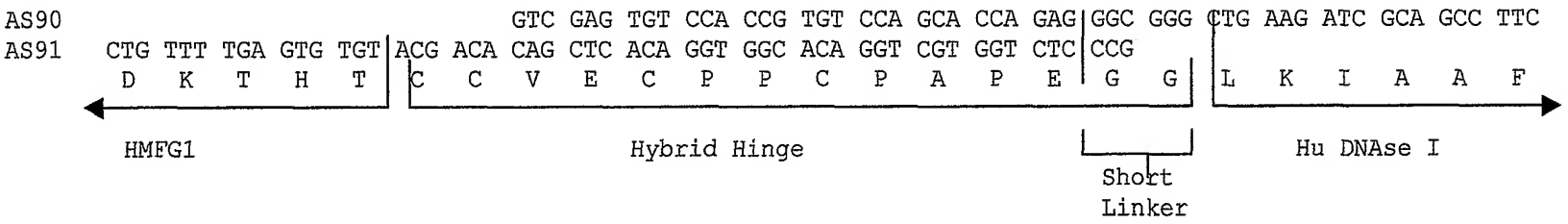


Fig. 4(B)

10/113

pAS23

```
LOCUS       PAS23.DNA     1554 bp    mRNA            PRI       06-MAR-1995
DEFINITION  Humanised HMFG1 Fab'2 fused to human DNase I (construct 1)
ACCESSION
NID
KEYWORDS    DNase I.
SOURCE      DNase I sequence is from assembled oligos (thus modified c/f
MHDNASE1.dna)
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
  AUTHORS   Shak,S., Capon,D.J., Hellmiss,R., Marsters,S.A. and Baker,C.L.
  TITLE     Recombinant human DNase I reduces the viscosity of cystic
fibrosis
            sputum
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 87 (23), 9188-9192 (1990)
  MEDLINE   91067672
BASE COUNT      344 a    468 c    434 g    308 t
ORIGIN

        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCCC ACCGTGCCCA GCACCTGAAG GGAGCGGCGG GCTGAAGATC
      781 GCAGCCTTCA ACATCCAGAC ATTTGGGGAG ACCAAGATGT CCAATGCCAC CCTCGTCAGC
      841 TACATTGTGC AGATCCTGAG CCGCTACGAC ATCGCCCTGG TCCAGGAGGT CAGAGACAGC
      901 CACCTGACTG CCGTGGGGAA GCTGCTGGAC AACCTCAATC AGGACGCACC AGACACCTAT
      961 CACTACGTGG TCAGTGAGCC ACTGGGACGG AACAGCTATA AGGAGCGCTA CCTGTTCGTG
     1021 TACAGGCCTG ACCAGGTGTC TGCGGTGGAC AGCTACTACT ACGATGATGG CTGCGAGCCC
     1081 TGCGGGAACG ACACCTTCAA CCGAGAGCCA GCCATTGTCA GGTTCTTCTC CCGGTTCACA
     1141 GAGGTCAGGG AGTTTGCCAT TGTTCCCCTG CATGCGGCCC CGGGGGACGC AGTAGCCGAG
     1201 ATCGACGCTC TCTATGACGT CTACCTGGAT GTCCAAGAGA AATGGGGCTT GGAGGACGTC
     1261 ATGTTGATGG GCGACTTCAA TGCGGGCTGC AGCTATGTGA GACCCTCCCA GTGGTCATCC
     1321 ATCCGCCTGT GGACAAGCCC CACCTTCCAG TGGCTGATCC CCGACAGCGC TGACACCACA
     1381 GCTACACCCA CGCACTGTGC CTATGACAGG ATCGTGGTTG CAGGGATGCT GCTCCGAGGG
     1441 GCCGTTGTTC CCGACTCGGC TCTTCCCTTT AACTTCCAGG CTGCCTATGG CCTGAGTGAC
     1501 CAACTGGCCC AAGCCATCAG TGACCACTAT CCAGTGGAGG TGATGCTGAA GTGA
//
```

Fig. 5(A)

11/113

```
LOCUS       FDDNASE23_    1554 BP SS-DNA             SYN       25-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
     frag            join(1..>720,<787..1554)
                     /note="1 to 1554 of 23.dna [Split]"
     frag            721..786
                     /note="1 to 66 of 23/27linker"
     frag            join(721..>735,<736..786)
                     /note="1 to 78 of 102linker [Split]"
BASE COUNT       344 A    466 C    435 G    309 T      0 OTHER
ORIGIN      -
        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGTCC ACCGTGTCCA GCACCAGAGG GGAGCGGCGG GCTGAAGATC
      781 GCAGCCTTCA ACATCCAGAC ATTTGGGGAG ACCAAGATGT CCAATGCCAC CCTCGTCAGC
      841 TACATTGTGC AGATCCTGAG CCGCTACGAC ATCGCCCTGG TCCAGGAGGT CAGAGACAGC
      901 CACCTGACTG CCGTGGGGAA GCTGCTGGAC AACCTCAATC AGGACGCACC AGACACCTAT
      961 CACTACGTGG TCAGTGAGCC ACTGGGACGG AACAGCTATA AGGAGCGCTA CCTGTTCGTG
     1021 TACAGGCCTG ACCAGGTGTC TGCGGTGGAC AGCTACTACT ACGATGATGG CTGCGAGCCC
     1081 TGCGGGAACG ACACCTTCAA CCGAGAGCCA GCCATTGTCA GGTTCTTCTC CCGGTTCACA
     1141 GAGGTCAGGG AGTTTGCCAT TGTTCCCCTG CATGCGGCCC CGGGGGACGC AGTAGCCGAG
     1201 ATCGACGCTC TCTATGACGT CTACCTGGAT GTCCAAGAGA AATGGGGCTT GGAGGACGTC
     1261 ATGTTGATGG GCGACTTCAA TGCGGGCTGC AGCTATGTGA GACCCTCCCA GTGGTCATCC
     1321 ATCCGCCTGT GGACAAGCCC CACCTTCCAG TGGCTGATCC CCGACAGCGC TGACACCACA
     1381 GCTACACCCA CGCACTGTGC CTATGACAGG ATCGTGGTTG CAGGGATGCT GCTCCGAGGG
     1441 GCCGTTGTTC CCGACTCGGC TCTTCCCTTT AACTTCCAGG CTGCCTATGG CCTGAGTGAC
     1501 CAACTGGCCC AAGCCATCAG TGACCACTAT CCAGTGGAGG TGATGCTGAA GTGA
//
```

Fig. 5(B)

12/113

```
LOCUS       FDDNASE23K   1563 BP SS-DNA             SYN       29-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
     frag            10..1563
                     /note="1 to 1554 of FdDNase23correct"
     frag            join(10..>729,<796..1563)
                     /note="1 to 1554 of 23.dna [Split]"
     frag            730..795
                     /note="1 to 66 of 23/27linker"
     frag            join(730..>744,<745..795)
                     /note="1 to 78 of 102linker [Split]"
BASE COUNT      345 A    472 C    437 G    309 T      0 OTHER
ORIGIN      -
        1 GCCGCCACCA TGGGATGGAG CTGTATCATC CTCTTCTTGG TAGCAACAGC TACAGGTGTC
       61 CACTCCCAGG TGCAGCTGGT GCAGTCTGGG GCAGAGGTGA AAAAGCCTGG GGCCTCAGTG
      121 AAGGTGTCCT GCAAGGCTTC TGGCTACACC TTCAGTGCCT ACTGGATAGA GTGGGTGCGC
      181 CAGGCTCCAG GAAAGGGCCT CGAGTGGGTC GGAGAGATTT TACCTGGAAG TAATAATTCT
      241 AGATACAATG AGAAGTTCAA GGGCCGAGTG ACAGTCACTA GAGACACATC CACAAACACA
      301 GCCTACATGG AGCTCAGCAG CCTGAGGTCT GAGGACACAG CCGTCTATTA CTGTGCAAGA
      361 TCCTACGACT TTGCCTGGTT TGCTTACTGG GGCCAAGGGA CTCTGGTCAC AGTCTCCTCA
      421 GCCTCCACCA AGGGCCCATC GGTCTTCCCC CTGGCACCCT CCTCCAAGAG CACCTCTGGG
      481 GGCACAGCGG CCCTGGGCTG CCTGGTCAAG GACTACTTCC CCGAACCGGT GACGGTGTCG
      541 TGGAACTCAG GCGCCCTGAC CAGCGGCGTG CACACCTTCC CGGCTGTCCT ACAGTCCTCA
      601 GGACTCTACT CCCTCAGCAG CGTGGTGACC GTGCCCTCCA GCAGCTTGGG CACCCAGACC
      661 TACATCTGCA ACGTGAATCA CAAGCCCAGC AACACCAAGG TGGACAAGAA AGTTGAGCCC
      721 AAATCTTGTG ACAAAACTCA CACATGTCCA CCGTGTCCAG CACCAGAGGG GAGCGGCGGG
      781 CTGAAGATCG CAGCCTTCAA CATCCAGACA TTTGGGGAGA CCAAGATGTC CAATGCCACC
      841 CTCGTCAGCT ACATTGTGCA GATCCTGAGC CGCTACGACA TCGCCCTGGT CCAGGAGGTC
      901 AGAGACAGCC ACCTGACTGC CGTGGGGAAG CTGCTGGACA ACCTCAATCA GGACGCACCA
      961 GACACCTATC ACTACGTGGT CAGTGAGCCA CTGGGACGGA ACAGCTATAA GGAGCGCTAC
     1021 CTGTTCGTGT ACAGGCCTGA CCAGGTGTCT GCGGTGGACA GCTACTACTA CGATGATGGC
     1081 TGCGAGCCCT GCGGGAACGA CACCTTCAAC CGAGAGCCAG CCATTGTCAG GTTCTTCTCC
     1141 CGGTTCACAG AGGTCAGGGA GTTTGCCATT GTTCCCCTGC ATGCGGCCCC GGGGGACGCA
     1201 GTAGCCGAGA TCGACGCTCT CTATGACGTC TACCTGGATG TCCAAGAGAA ATGGGGCTTG
     1261 GAGGACGTCA TGTTGATGGG CGACTTCAAT GCGGGCTGCA GCTATGTGAG ACCCTCCCAG
     1321 TGGTCATCCA TCCGCCTGTG GACAAGCCCC ACCTTCCAGT GGCTGATCCC CGACAGCGCT
     1381 GACACCACAG CTACACCCAC GCACTGTGCC TATGACAGGA TCGTGGTTGC AGGGATGCTG
     1441 CTCCGAGGGG CCGTTGTTCC CGACTCGGCT CTTCCCTTTA ACTTCCAGGC TGCCTATGGC
     1501 CTGAGTGACC AACTGGCCCA AGCCATCAGT GACCACTATC CAGTGGAGGT GATGCTGAAG
     1561 TGA
//
```

Fig. 5(C)

```
                9         18        27        36        45        54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63        72        81        90        99       108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117       126       135       144       153       162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171       180       189       198       207       216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225       234       243       252       261       270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279       288       297       306       315       324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333       342       351       360       369       378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387       396       405       414       423       432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441       450       459       468       477       486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495       504       513       522       531       540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549       558       567       576       585       594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G
```


Fig. 5(D)

(Sheet 1 of 3)

```
         603         612         621         630         639         648
CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

         657         666         675         684         693         702
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

         711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC CCA CCG TGC CCA GCA CCT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   P   P   C   P   A   P

         765         774         783         792         801         810
GAA GGG AGC GGC GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT GGG GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   G   S   G   G   L   K   I   A   A   F   N   I   Q   T   F   G   E

         819         828         837         846         855         864
ACC AAG ATG TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG AGC CGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   K   M   S   N   A   T   L   V   S   Y   I   V   Q   I   L   S   R

         873         882         891         900         909         918
TAC GAC ATC GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC GTG GGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Y   D   I   A   L   V   Q   E   V   R   D   S   H   L   T   A   V   G

         927         936         945         954         963         972
AAG CTG CTG GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC GTG GTC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 K   L   L   D   N   L   N   Q   D   A   P   D   T   Y   H   Y   V   V

         981         990         999        1008        1017        1026
AGT GAG CCA CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG TAC AGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   E   P   L   G   R   N   S   Y   K   E   R   Y   L   F   V   Y   R

        1035        1044        1053        1062        1071        1080
CCT GAC CAG GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC GAG CCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   D   Q   V   S   A   V   D   S   Y   Y   Y   D   D   G   C   E   P

        1089        1098        1107        1116        1125        1134
TGC GGG AAC GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC TCC CGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 C   G   N   D   T   F   N   R   E   P   A   I   V   R   F   F   S   R

        1143        1152        1161        1170        1179        1188
TTC ACA GAG GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GCC CCG GGG GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 F   T   E   V   R   E   F   A   I   V   P   L   H   A   A   P   G   D

        1197        1206        1215        1224        1233        1242
```

Fig. 5(D)

(Sheet 2 of 3)

```
GCA GTA GCC GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA GAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   V   A   E   I   D   A   L   Y   D   V   Y   L   D   V   Q   E   K

      1251        1260        1269        1278        1287        1296
TGG GGC TTG GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC AGC TAT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 W   G   L   E   D   V   M   L   M   G   D   F   N   A   G   C   S   Y

      1305        1314        1323        1332        1341        1350
GTG AGA CCC TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC TTC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   R   P   S   Q   W   S   S   I   R   L   W   T   S   P   T   F   Q

      1359        1368        1377        1386        1395        1404
TGG CTG ATC CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT GCC TAT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 W   L   I   P   D   S   A   D   T   T   A   T   P   T   H   C   A   Y

      1413        1422        1431        1440        1449        1458
GAC AGG ATC GTG GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT GTT CCC GAC TCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   R   I   V   V   A   G   M   L   L   R   G   A   V   V   P   D   S

      1467        1476        1485        1494        1503        1512
GCT CTT CCC TTT AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC CAA CTG GCC CAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   L   P   F   N   F   Q   A   A   Y   G   L   S   D   Q   L   A   Q

      1521        1530        1539        1548
GCC ATC AGT GAC CAC TAT CCA GTG GAG GTG ATG CTG AAG TGA 3'
--- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   I   S   D   H   Y   P   V   E   V   M   L   K   *
```

Fig. 5(D)

(Sheet 3 of 3)

16/113

pAS27

LOCUS PAS27.DNA 1584 bp mRNA PRI 06-MAR-1995
DEFINITION Humanised HMFG1 Fab'2 fused to human DNase I with SV40 NLS(construct 1)
ACCESSION
NID
KEYWORDS DNase I.
SOURCE DNase I sequence is from assembled oligos (thus modified c/f MHDNASE1.dna)
ORGANISM Homo sapiens
Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata; Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
AUTHORS Shak,S., Capon,D.J., Hellmiss,R., Marsters,S.A. and Baker,C.L.
TITLE Recombinant human DNase I reduces the viscosity of cystic fibrosis sputum
JOURNAL Proc. Natl. Acad. Sci. U.S.A. 87 (23), 9188-9192 (1990)
MEDLINE 91067672
BASE COUNT 354 a 474 c 446 g 310 t
ORIGIN

```
       1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
      61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
     121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
     181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
     241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
     301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
     361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
     421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
     481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
     541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
     601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
     661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
     721 GACAAAACTC ACACATGCCC ACCGTGCCCA GCACCTGAAG GGAGCGGCGG GCTGAAGATC
     781 GCAGCCTTCA ACATCCAGAC ATTTGGGGAG ACCAAGATGT CCAATGCCAC CCTCGTCAGC
     841 TACATTGTGC AGATCCTGAG CCGCTACGAC ATCGCCCTGG TCCAGGAGGT CAGAGACAGC
     901 CACCTGACTG CCGTGGGGAA GCTGCTGGAC AACCTCAATC AGGACGCACC AGACACCTAT
     961 CACTACGTGG TCAGTGAGCC ACTGGGACGG AACAGCTATA AGGAGCGCTA CCTGTTCGTG
    1021 TACAGGCCTG ACCAGGTGTC TGCGGTGGAC AGCTACTACT ACGATGATGG CTGCGAGCCC
    1081 TGCGGGAACG ACACCTTCAA CCGAGAGCCA GCCATTGTCA GGTTCTTCTC CCGGTTCACA
    1141 GAGGTCAGGG AGTTTGCCAT TGTTCCCCTG CATGCGGCCC CGGGGGACGC AGTAGCCGAG
    1201 ATCGACGCTC TCTATGACGT CTACCTGGAT GTCCAAGAGA AATGGGGCTT GGAGGACGTC
    1261 ATGTTGATGG GCGACTTCAA TGCGGGCTGC AGCTATGTGA GACCCTCCCA GTGGTCATCC
    1321 ATCCGCCTGT GGACAAGCCC CACCTTCCAG TGGCTGATCC CCGACAGCGC TGACACCACA
    1381 GCTACACCCA CGCACTGTGC CTATGACAGG ATCGTGGTTG CAGGGATGCT GCTCCGAGGG
    1441 GCCGTTGTTC CCGACTCGGC TCTTCCCTTT AACTTCCAGG CTGCCTATGG CCTGAGTGAC
    1501 CAACTGGCCC AAGCCATCAG TGACCACTAT CCAGTGGAGG TGATGCTGAA GGGGGGCGGA
    1561 CCCAAAAAGA AGCGCAAGGT TTGA
//
```

Fig. 6(A)

17/113

```
LOCUS       FDDNASE27_   1584 BP SS-DNA             SYN       25-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
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                     /note="1 to 1584 of 27.dna [Split]"
     frag            721..786
                     /note="1 to 66 of 23/27linker"
     frag            join(721..>735,<736..786)
                     /note="1 to 78 of 102linker [Split]"
BASE COUNT       354 A    472 C    447 G    311 T      0 OTHER
ORIGIN      -
        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGTCC ACCGTGTCCA GCACCAGAGG GGAGCGGCGG GCTGAAGATC
      781 GCAGCCTTCA ACATCCAGAC ATTTGGGGAG ACCAAGATGT CCAATGCCAC CCTCGTCAGC
      841 TACATTGTGC AGATCCTGAG CCGCTACGAC ATCGCCCTGG TCCAGGAGGT CAGAGACAGC
      901 CACCTGACTG CCGTGGGGAA GCTGCTGGAC AACCTCAATC AGGACGCACC AGACACCTAT
      961 CACTACGTGG TCAGTGAGCC ACTGGGACGG AACAGCTATA AGGAGCGCTA CCTGTTCGTG
     1021 TACAGGCCTG ACCAGGTGTC TGCGGTGGAC AGCTACTACT ACGATGATGG CTGCGAGCCC
     1081 TGCGGGAACG ACACCTTCAA CCGAGAGCCA GCCATTGTCA GGTTCTTCTC CCGGTTCACA
     1141 GAGGTCAGGG AGTTTGCCAT TGTTCCCCTG CATGCGGCCC CGGGGGACGC AGTAGCCGAG
     1201 ATCGACGCTC TCTATGACGT CTACCTGGAT GTCCAAGAGA AATGGGGCTT GGAGGACGTC
     1261 ATGTTGATGG GCGACTTCAA TGCGGGCTGC AGCTATGTGA GACCCTCCCA GTGGTCATCC
     1321 ATCCGCCTGT GGACAAGCCC CACCTTCCAG TGGCTGATCC CCGACAGCGC TGACACCACA
     1381 GCTACACCCA CGCACTGTGC CTATGACAGG ATCGTGGTTG CAGGGATGCT GCTCCGAGGG
     1441 GCCGTTGTTC CCGACTCGGC TCTTCCCTTT AACTTCCAGG CTGCCTATGG CCTGAGTGAC
     1501 CAACTGGCCC AAGCCATCAG TGACCACTAT CCAGTGGAGG TGATGCTGAA GGGGGGCGGA
     1561 CCCAAAAAGA AGCGCAAGGT TTGA
//
```

Fig. 6(B)

18/113

```
LOCUS       FDDNASE27K   1593 BP SS-DNA             SYN       29-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
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                     /note="1 to 1584 of FdDNase27correct"
     frag            join(10..>729,<796..1593)
                     /note="1 to 1584 of 27.dna [Split]"
     frag            730..795
                     /note="1 to 66 of 23/27linker"
     frag            join(730..>744,<745..795)
                     /note="1 to 78 of 102linker [Split]"
BASE COUNT      355 A    478 C    449 G    311 T      0 OTHER
ORIGIN      -
        1 GCCGCCACCA TGGGATGGAG CTGTATCATC CTCTTCTTGG TAGCAACAGC TACAGGTGTC
       61 CACTCCCAGG TGCAGCTGGT GCAGTCTGGG GCAGAGGTGA AAAAGCCTGG GGCCTCAGTG
      121 AAGGTGTCCT GCAAGGCTTC TGGCTACACC TTCAGTGCCT ACTGGATAGA GTGGGTGCGC
      181 CAGGCTCCAG GAAAGGGCCT CGAGTGGGTC GGAGAGATTT TACCTGGAAG TAATAATTCT
      241 AGATACAATG AGAAGTTCAA GGGCCGAGTG ACAGTCACTA GAGACACATC CACAAACACA
      301 GCCTACATGG AGCTCAGCAG CCTGAGGTCT GAGGACACAG CCGTCTATTA CTGTGCAAGA
      361 TCCTACGACT TTGCCTGGTT TGCTTACTGG GGCCAAGGGA CTCTGGTCAC AGTCTCCTCA
      421 GCCTCCACCA AGGGCCCATC GGTCTTCCCC CTGGCACCCT CCTCCAAGAG CACCTCTGGG
      481 GGCACAGCGG CCCTGGGCTG CCTGGTCAAG GACTACTTCC CCGAACCGGT GACGGTGTCG
      541 TGGAACTCAG GCGCCCTGAC CAGCGGCGTG CACACCTTCC CGGCTGTCCT ACAGTCCTCA
      601 GGACTCTACT CCCTCAGCAG CGTGGTGACC GTGCCCTCCA GCAGCTTGGG CACCCAGACC
      661 TACATCTGCA ACGTGAATCA CAAGCCCAGC AACACCAAGG TGGACAAGAA AGTTGAGCCC
      721 AAATCTTGTG ACAAAACTCA CACATGTCCA CCGTGTCCAG CACCAGAGGG GAGCGGCGGG
      781 CTGAAGATCG CAGCCTTCAA CATCCAGACA TTTGGGGAGA CCAAGATGTC CAATGCCACC
      841 CTCGTCAGCT ACATTGTGCA GATCCTGAGC CGCTACGACA TCGCCCTGGT CCAGGAGGTC
      901 AGAGACAGCC ACCTGACTGC CGTGGGGAAG CTGCTGGACA ACCTCAATCA GGACGCACCA
      961 GACACCTATC ACTACGTGGT CAGTGAGCCA CTGGGACGGA ACAGCTATAA GGAGCGCTAC
     1021 CTGTTCGTGT ACAGGCCTGA CCAGGTGTCT GCGGTGGACA GCTACTACTA CGATGATGGC
     1081 TGCGAGCCCT GCGGGAACGA CACCTTCAAC CGAGAGCCAG CCATTGTCAG GTTCTTCTCC
     1141 CGGTTCACAG AGGTCAGGGA GTTTGCCATT GTTCCCCTGC ATGCGGCCCC GGGGGACGCA
     1201 GTAGCCGAGA TCGACGCTCT CTATGACGTC TACCTGGATG TCCAAGAGAA ATGGGGCTTG
     1261 GAGGACGTCA TGTTGATGGG CGACTTCAAT GCGGGCTGCA GCTATGTGAG ACCCTCCCAG
     1321 TGGTCATCCA TCCGCCTGTG GACAAGCCCC ACCTTCCAGT GGCTGATCCC CGACAGCGCT
     1381 GACACCACAG CTACACCCAC GCACTGTGCC TATGACAGGA TCGTGGTTGC AGGGATGCTG
     1441 CTCCGAGGGG CCGTTGTTCC CGACTCGGCT CTTCCCTTTA ACTTCCAGGC TGCCTATGGC
     1501 CTGAGTGACC AACTGGCCCA AGCCATCAGT GACCACTATC CAGTGGAGGT GATGCTGAAG
     1561 GGGGGCGGAC CCAAAAAGAA GCGCAAGGTT TGA
//
```

Fig. 6(C)

19/113

```
                9          18          27          36          45          54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63          72          81          90          99         108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117         126         135         144         153         162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171         180         189         198         207         216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225         234         243         252         261         270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279         288         297         306         315         324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333         342         351         360         369         378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387         396         405         414         423         432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441         450         459         468         477         486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495         504         513         522         531         540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549         558         567         576         585         594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G
```


Fig. 6(D)

(Sheet 1 of 3)

20/113

```
        603         612         621         630         639         648
CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

        657         666         675         684         693         702
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC CCA CCG TGC CCA GCA CCT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   P   P   C   P   A   P

        765         774         783         792         801         810
GAA GGG AGC GGC GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT GGG GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   G   S   G   G   L   K   I   A   A   F   N   I   Q   T   F   G   E

        819         828         837         846         855         864
ACC AAG ATG TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG AGC CGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   K   M   S   N   A   T   L   V   S   Y   I   V   Q   I   L   S   R

        873         882         891         900         909         918
TAC GAC ATC GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC GTG GGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Y   D   I   A   L   V   Q   E   V   R   D   S   H   L   T   A   V   G

        927         936         945         954         963         972
AAG CTG CTG GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC GTG GTC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 K   L   L   D   N   L   N   Q   D   A   P   D   T   Y   H   Y   V   V

        981         990         999        1008        1017        1026
AGT GAG CCA CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG TAC AGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   E   P   L   G   R   N   S   Y   K   E   R   Y   L   F   V   Y   R

       1035        1044        1053        1062        1071        1080
CCT GAC CAG GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC GAG CCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   D   Q   V   S   A   V   D   S   Y   Y   Y   D   D   G   C   E   P

       1089        1098        1107        1116        1125        1134
TGC GGG AAC GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC TCC CGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 C   G   N   D   T   F   N   R   E   P   A   I   V   R   F   F   S   R

       1143        1152        1161        1170        1179        1188
TTC ACA GAG GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GCC CCG GGG GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 F   T   E   V   R   E   F   A   I   V   P   L   H   A   A   P   G   D

       1197        1206        1215        1224        1233        1242
```

Fig. 6(D)

(Sheet 2 of 3)

21/113

```
GCA GTA GCC GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA GAG AAA
---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---
 A    V    A    E    I    D    A    L    Y    D    V    Y    L    D    V    Q    E    K

       1251         1260         1269         1278         1287         1296
TGG GGC TTG GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC AGC TAT
---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---
 W    G    L    E    D    V    M    L    M    G    D    F    N    A    G    C    S    Y

       1305         1314         1323         1332         1341         1350
GTG AGA CCC TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC TTC CAG
---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---
 V    R    P    S    Q    W    S    S    I    R    L    W    T    S    P    T    F    Q

       1359         1368         1377         1386         1395         1404
TGG CTG ATC CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT GCC TAT
---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---
 W    L    I    P    D    S    A    D    T    T    A    T    P    T    H    C    A    Y

       1413         1422         1431         1440         1449         1458
GAC AGG ATC GTG GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT GTT CCC GAC TCG
---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---
 D    R    I    V    V    A    G    M    L    L    R    G    A    V    V    P    D    S

       1467         1476         1485         1494         1503         1512
GCT CTT CCC TTT AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC CAA CTG GCC CAA
---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---
 A    L    P    F    N    F    Q    A    A    Y    G    L    S    D    Q    L    A    Q

       1521         1530         1539         1548         1557         1566
GCC ATC AGT GAC CAC TAT CCA GTG GAG GTG ATG CTG AAG GGG GGC GGA CCC AAA
---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---  ---
 A    I    S    D    H    Y    P    V    E    V    M    L    K    G    G    G    P    K

       1575         1584
AAG AAG CGC AAG GTT TGA 3'
---  ---  ---  ---  ---  ---
 K    K    R    K    V    *
```

Fig. 6D

(Sheet 3 of 3)

22/113

pAS34

```
LOCUS       PAS34.DNA    2196 bp        2196 bp  2196 bp  DNA   14-AUG-1998
DEFINITION  HUMANISED HMFG1 heavy chain fused to human DNAse construct 34
DEFINITION  Clone 16.4.2 (same as hcdnase1.dna template file)
REFERENCE
  AUTHORS   VERHOEYEN ET AL
  TITLE     CONSTRUCTION OF RESHAPED HMFG1 etc
  JOURNAL   IMMUNOL. (1993):78, 364-370
COMMENT     Human DNAse sequence is modified as a result of oligo assembly
(mhdnase.dna)
COMMENT     The fusion was made using overlapping oligos AS79 and AS80
FEATURES    AA RESIDUE 235 HAS NOT BEEN CHANGED TO KABAT (I.E. V TO A)
FEATURES    Residue 963 is G > T leading to silent mutation in all clones
  SITES     Note
BASE COUNT      501 a    677 c    607 g    411 t
ORIGIN           ?

        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCCC ACCGTGCCCA GCACCTGAAC TCCTGGGGGG ACCGTCAGTC
      781 TTCCTCTTCC CCCCAAAACC CAAGGACACC CTCATGATCT CCCGGACCCC TGAGGTCACA
      841 TGCGTGGTGG TGGACGTGAG CCACGAAGAC CCTGAGGTCA AGTTCAACTG GTACGTGGAC
      901 GGCGTGGAGG TGCATAATGC CAAGACAAAG CCGCGGGAGG AGCAGTACAA CAGCACGTAC
      961 CGTGTGGTCA GCGTCCTCAC CGTCCTGCAC CAGGACTGGC TGAATGGCAA GGAGTACAAG
     1021 TGCAAGGTCT CCAACAAAGC CCTCCCAGCC CCCATCGAGA AAACCATCTC CAAAGCCAAA
     1081 GGGCAGCCCC GAGAACCACA GGTGTACACC CTGCCCCCAT CCCGGGATGA GCTGACCAAG
     1141 AACCAGGTCA GCCTGACCTG CCTGGTCAAA GGCTTCTATC CCAGCGACAT CGCCGTGGAG
     1201 TGGGAGAGCA ATGGGCAGCC GGAGAACAAC TACAAGACCA CGCCTCCCGT GCTGGACTCC
     1261 GACGGCTCCT TCTTCCTCTA CAGCAAGCTC ACCGTGGACA AGAGCAGGTG GCAGCAGGGG
     1321 AACGTCTTCT CATGCTCCGT GATGCATGAG GCTCTGCACA ACCACTACAC GCAGAAGAGC
     1381 CTCTCCCTGT CTCCGGGTAA AGGGAGCGGC GGGCTGAAGA TCGCAGCCTT CAACATCCAG
     1441 ACATTTGGGG AGACCAAGAT GTCCAATGCC ACCCTCGTCA GCTACATTGT GCAGATCCTG
     1501 AGCCGCTACG ACATCGCCCT GGTCCAGGAG GTCAGAGACA GCCACCTGAC TGCCGTGGGG
     1561 AAGCTGCTGG ACAACCTCAA TCAGGACGCA CCAGACACCT ATCACTACGT GGTCAGTGAG
     1621 CCACTGGGAC GGAACAGCTA TAAGGAGCGC TACCTGTTCG TGTACAGGCC TGACCAGGTG
     1681 TCTGCGGTGG ACAGCTACTA CTACGATGAT GGCTGCGAGC CCTGCGGGAA CGACACCTTC
     1741 AACCGAGAGC CAGCCATTGT CAGGTTCTTC TCCCGGTTCA CAGAGGTCAG GGAGTTTGCC
     1801 ATTGTTCCCC TGCATGCGGC CCCGGGGGAC GCAGTAGCCG AGATCGACGC TCTCTATGAC
     1861 GTCTACCTGG ATGTCCAAGA GAAATGGGGC TTGGAGGACG TCATGTTGAT GGGCGACTTC
     1921 AATGCGGGCT GCAGCTATGT GAGACCCTCC CAGTGGTCAT CCATCCGCCT GTGGACAAGC
     1981 CCCACCTTCC AGTGGCTGAT CCCCGACAGC GCTGACACCA CAGCTACACC CACGCACTGT
     2041 GCCTATGACA GGATCGTGGT TGCAGGGATG CTGCTCCGAG GGGCCGTTGT TCCCGACTCG
     2101 GCTCTTCCCT TTAACTTCCA GGCTGCCTAT GGCCTGAGTG ACCAACTGGC CCAAGCCATC
     2161 AGTGACCACT ATCCAGTGGA GGTGATGCTG AAGTGA
//
```

Fig. 7(A)

```
                9          18         27         36         45         54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63         72         81         90         99        108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117        126        135        144        153        162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171        180        189        198        207        216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225        234        243        252        261        270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279        288        297        306        315        324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333        342        351        360        369        378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387        396        405        414        423        432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441        450        459        468        477        486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495        504        513        522        531        540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549        558        567        576        585        594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G

              603        612        621        630        639        648
    CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

              657        666        675        684        693        702
```

Fig. 7(B)
(Sheet 1 of 4)

```
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC CCA CCG TGC CCA GCA CCT
 V   E   P   K   S   C   D   K   T   H   T   C   P   P   C   P   A   P

        765         774         783         792         801         810
GAA CTC CTG GGG GGA CCG TCA GTC TTC CTC TTC CCC CCA AAA CCC AAG GAC ACC
 E   L   L   G   G   P   S   V   F   L   F   P   P   K   P   K   D   T

        819         828         837         846         855         864
CTC ATG ATC TCC CGG ACC CCT GAG GTC ACA TGC GTG GTG GTG GAC GTG AGC CAC
 L   M   I   S   R   T   P   E   V   T   C   V   V   V   D   V   S   H

        873         882         891         900         909         918
GAA GAC CCT GAG GTC AAG TTC AAC TGG TAC GTG GAC GGC GTG GAG GTG CAT AAT
 E   D   P   E   V   K   F   N   W   Y   V   D   G   V   E   V   H   N

        927         936         945         954         963         972
GCC AAG ACA AAG CCG CGG GAG GAG CAG TAC AAC AGC ACG TAC CGT GTG GTC AGC
 A   K   T   K   P   R   E   E   Q   Y   N   S   T   Y   R   V   V   S

        981         990         999        1008        1017        1026
GTC CTC ACC GTC CTG CAC CAG GAC TGG CTG AAT GGC AAG GAG TAC AAG TGC AAG
 V   L   T   V   L   H   Q   D   W   L   N   G   K   E   Y   K   C   K

       1035        1044        1053        1062        1071        1080
GTC TCC AAC AAA GCC CTC CCA GCC CCC ATC GAG AAA ACC ATC TCC AAA GCC AAA
 V   S   N   K   A   L   P   A   P   I   E   K   T   I   S   K   A   K

       1089        1098        1107        1116        1125        1134
GGG CAG CCC CGA GAA CCA CAG GTG TAC ACC CTG CCC CCA TCC CGG GAT GAG CTG
 G   Q   P   R   E   P   Q   V   Y   T   L   P   P   S   R   D   E   L

       1143        1152        1161        1170        1179        1188
ACC AAG AAC CAG GTC AGC CTG ACC TGC CTG GTC AAA GGC TTC TAT CCC AGC GAC
 T   K   N   Q   V   S   L   T   C   L   V   K   G   F   Y   P   S   D

       1197        1206        1215        1224        1233        1242
ATC GCC GTG GAG TGG GAG AGC AAT GGG CAG CCG GAG AAC AAC TAC AAG ACC ACG
 I   A   V   E   W   E   S   N   G   Q   P   E   N   N   Y   K   T   T

       1251        1260        1269        1278        1287        1296
CCT CCC GTG CTG GAC TCC GAC GGC TCC TTC TTC CTC TAC AGC AAG CTC ACC GTG
 P   P   V   L   D   S   D   G   S   F   F   L   Y   S   K   L   T   V

       1305        1314        1323        1332        1341        1350
GAC AAG AGC AGG TGG CAG CAG GGG AAC GTC TTC TCA TGC TCC GTG ATG CAT GAG
 D   K   S   R   W   Q   Q   G   N   V   F   S   C   S   V   M   H   E
```

Fig. 7(B)

(Sheet 2 of 4)

```
      1359       1368       1377       1386       1395       1404
GCT CTG CAC AAC CAC TAC ACG CAG AAG AGC CTC TCC CTG TCT CCG GGT AAA GGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   L   H   N   H   Y   T   Q   K   S   L   S   L   S   P   G   K   G

      1413       1422       1431       1440       1449       1458
AGC GGC GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT GGG GAG ACC AAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   G   G   L   K   I   A   A   F   N   I   Q   T   F   G   E   T   K

      1467       1476       1485       1494       1503       1512
ATG TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG AGC CGC TAC GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 M   S   N   A   T   L   V   S   Y   I   V   Q   I   L   S   R   Y   D

      1521       1530       1539       1548       1557       1566
ATC GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC GTG GGG AAG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   A   L   V   Q   E   V   R   D   S   H   L   T   A   V   G   K   L

      1575       1584       1593       1602       1611       1620
CTG GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC GTG GTC AGT GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   D   N   L   N   Q   D   A   P   D   T   Y   H   Y   V   V   S   E

      1629       1638       1647       1656       1665       1674
CCA CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG TAC AGG CCT GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   L   G   R   N   S   Y   K   E   R   Y   L   F   V   Y   R   P   D

      1683       1692       1701       1710       1719       1728
CAG GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC GAG CCC TGC GGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Q   V   S   A   V   D   S   Y   Y   Y   D   D   G   C   E   P   C   G

      1737       1746       1755       1764       1773       1782
AAC GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC TCC CGG TTC ACA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 N   D   T   F   N   R   E   P   A   I   V   R   F   F   S   R   F   T

      1791       1800       1809       1818       1827       1836
GAG GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GCC CCG GGG GAC GCA GTA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   V   R   E   F   A   I   V   P   L   H   A   A   P   G   D   A   V

      1845       1854       1863       1872       1881       1890
GCC GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA GAG AAA TGG GGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   E   I   D   A   L   Y   D   V   Y   L   D   V   Q   E   K   W   G

      1899       1908       1917       1926       1935       1944
TTG GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC AGC TAT GTG AGA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   E   D   V   M   L   M   G   D   F   N   A   G   C   S   Y   V   R

      1953       1962       1971       1980       1989       1998
CCC TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC TTC CAG TGG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   S   Q   W   S   S   I   R   L   W   T   S   P   T   F   Q   W   L

      2007       2016       2025       2034       2043       2052
ATC CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT GCC TAT GAC AGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   P   D   S   A   D   T   T   A   T   P   T   H   C   A   Y   D   R
```

Fig. 7(B)

(Sheet 3 of 4)

26/113

```
      2061        2070        2079        2088        2097        2106
ATC GTG GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT GTT CCC GAC TCG GCT CTT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   V   V   A   G   M   L   L   R   G   A   V   V   P   D   S   A   L

      2115        2124        2133        2142        2151        2160
CCC TTT AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC CAA CTG GCC CAA GCC ATC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   F   N   F   Q   A   A   Y   G   L   S   D   Q   L   A   Q   A   I

      2169        2178        2187        2196
AGT GAC CAC TAT CCA GTG GAG GTG ATG CTG AAG TGA 3'
--- --- --- --- --- --- --- --- --- --- --- ---
 S   D   H   Y   P   V   E   V   M   L   K   *
```

Fig. 7(B)

(Sheet 4 of 4)

27/113

pAS35

```
LOCUS       PAS35.DNA    2193 bp   2193 bp    DNA              14-AUG-1998
DEFINITION  HUMANISED HMFG1 heavy chain fused to human DNAse construct 35
DEFINITION  Clone 17.12.1 with silent K to K mutation (1398 A > G)
REFERENCE
  AUTHORS   VERHOEYEN ET AL
  TITLE     CONSTRUCTION OF RESHAPED HMFG1 etc
  JOURNAL   IMMUNOL. (1993):78, 364-370
COMMENT     Human DNAse sequence is modified as a result of oligo assembly
(mhdnase.dna)
COMMENT     The fusion was made using overlapping oligos AS81 and AS82
FEATURES    AA RESIDUE 235 HAS NOT BEEN CHANGED TO KABAT (I.E. V TO A)
FEATURES    Residue 963 is G > T leading to silent mutation in all clones
FEATURES    In 17.12.1 residue 1398 is A > G (silent K to K mutation)
  SITES     Note
BASE COUNT      500 a    677 c    606 g    410 t
ORIGIN           ?

        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCCC ACCGTGCCCA GCACCTGAAC TCCTGGGGGG ACCGTCAGTC
      781 TTCCTCTTCC CCCCAAAACC CAAGGACACC CTCATGATCT CCCGGACCCC TGAGGTCACA
      841 TGCGTGGTGG TGGACGTGAG CCACGAAGAC CCTGAGGTCA AGTTCAACTG GTACGTGGAC
      901 GGCGTGGAGG TGCATAATGC CAAGACAAAG CCGCGGGAGG AGCAGTACAA CAGCACGTAC
      961 CGTGTGGTCA GCGTCCTCAC CGTCCTGCAC CAGGACTGGC TGAATGGCAA GGAGTACAAG
     1021 TGCAAGGTCT CCAACAAAGC CCTCCCAGCC CCCATCGAGA AAACCATCTC CAAAGCCAAA
     1081 GGGCAGCCCC GAGAACCACA GGTGTACACC CTGCCCCCAT CCCGGGATGA GCTGACCAAG
     1141 AACCAGGTCA GCCTGACCTG CCTGGTCAAA GGCTTCTATC CCAGCGACAT CGCCGTGGAG
     1201 TGGGAGAGCA ATGGGCAGCC GGAGAACAAC TACAAGACCA CGCCTCCCGT GCTGGACTCC
     1261 GACGGCTCCT TCTTCCTCTA CAGCAAGCTC ACCGTGGACA AGAGCAGGTG GCAGCAGGGG
     1321 AACGTCTTCT CATGCTCCGT GATGCATGAG GCTCTGCACA ACCACTACAC GCAGAAGAGC
     1381 CTCTCCCTGT CTCCGAAgGG GAGCGGCGGG CTGAAGATCG CAGCCTTCAA CATCCAGACA
     1441 TTTGGGGAGA CCAAGATGTC CAATGCCACC CTCGTCAGCT ACATTGTGCA GATCCTGAGC
     1501 CGCTACGACA TCGCCCTGGT CCAGGAGGTC AGAGACAGCC ACCTGACTGC CGTGGGGAAG
     1561 CTGCTGGACA ACCTCAATCA GGACGCACCA GACACCTATC ACTACGTGGT CAGTGAGCCA
     1621 CTGGGACGGA ACAGCTATAA GGAGCGCTAC CTGTTCGTGT ACAGGCCTGA CCAGGTGTCT
     1681 GCGGTGGACA GCTACTACTA CGATGATGGC TGCGAGCCCT GCGGGAACGA CACCTTCAAC
     1741 CGAGAGCCAG CCATTGTCAG GTTCTTCTCC CGGTTCACAG AGGTCAGGGA GTTTGCCATT
     1801 GTTCCCCTGC ATGCGGCCCC GGGGGACGCA GTAGCCGAGA TCGACGCTCT CTATGACGTC
     1861 TACCTGGATG TCCAAGAGAA ATGGGGCTTG GAGGACGTCA TGTTGATGGG CGACTTCAAT
     1921 GCGGGCTGCA GCTATGTGAG ACCCTCCCAG TGGTCATCCA TCCGCCTGTG GACAAGCCCC
     1981 ACCTTCCAGT GGCTGATCCC CGACAGCGCT GACACCACAG CTACACCCAC GCACTGTGCC
     2041 TATGACAGGA TCGTGGTTGC AGGGATGCTG CTCCGAGGGG CCGTTGTTCC CGACTCGGCT
     2101 CTTCCCTTTA ACTTCCAGGC TGCCTATGGC CTGAGTGACC AACTGGCCCA AGCCATCAGT
     2161 GACCACTATC CAGTGGAGGT GATGCTGAAG TGA
//
```

Fig. 8(A)

28/113

```
                9          18         27         36         45         54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63         72         81         90         99        108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117        126        135        144        153        162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171        180        189        198        207        216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225        234        243        252        261        270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279        288        297        306        315        324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333        342        351        360        369        378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387        396        405        414        423        432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441        450        459        468        477        486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495        504        513        522        531        540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549        558        567        576        585        594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G

              603        612        621        630        639        648
    CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

              657        666        675        684        693        702
```

Fig. 8(B)

(Sheet 1 of 4)

```
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC CCA CCG TGC CCA GCA CCT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   P   P   C   P   A   P

        765         774         783         792         801         810
GAA CTC CTG GGG GGA CCG TCA GTC TTC CTC TTC CCC CCA AAA CCC AAG GAC ACC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   L   L   G   G   P   S   V   F   L   F   P   P   K   P   K   D   T

        819         828         837         846         855         864
CTC ATG ATC TCC CGG ACC CCT GAG GTC ACA TGC GTG GTG GTG GAC GTG AGC CAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   M   I   S   R   T   P   E   V   T   C   V   V   V   D   V   S   H

        873         882         891         900         909         918
GAA GAC CCT GAG GTC AAG TTC AAC TGG TAC GTG GAC GGC GTG GAG GTG CAT AAT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   D   P   E   V   K   F   N   W   Y   V   D   G   V   E   V   H   N

        927         936         945         954         963         972
GCC AAG ACA AAG CCG CGG GAG GAG CAG TAC AAC AGC ACG TAC CGT GTG GTC AGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   K   T   K   P   R   E   E   Q   Y   N   S   T   Y   R   V   V   S

        981         990         999        1008        1017        1026
GTC CTC ACC GTC CTG CAC CAG GAC TGG CTG AAT GGC AAG GAG TAC AAG TGC AAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   L   T   V   L   H   Q   D   W   L   N   G   K   E   Y   K   C   K

       1035        1044        1053        1062        1071        1080
GTC TCC AAC AAA GCC CTC CCA GCC CCC ATC GAG AAA ACC ATC TCC AAA GCC AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   S   N   K   A   L   P   A   P   I   E   K   T   I   S   K   A   K

       1089        1098        1107        1116        1125        1134
GGG CAG CCC CGA GAA CCA CAG GTG TAC ACC CTG CCC CCA TCC CGG GAT GAG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   Q   P   R   E   P   Q   V   Y   T   L   P   P   S   R   D   E   L

       1143        1152        1161        1170        1179        1188
ACC AAG AAC CAG GTC AGC CTG ACC TGC CTG GTC AAA GGC TTC TAT CCC AGC GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   K   N   Q   V   S   L   T   C   L   V   K   G   F   Y   P   S   D

       1197        1206        1215        1224        1233        1242
ATC GCC GTG GAG TGG GAG AGC AAT GGG CAG CCG GAG AAC AAC TAC AAG ACC ACG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   A   V   E   W   E   S   N   G   Q   P   E   N   N   Y   K   T   T

       1251        1260        1269        1278        1287        1296
CCT CCC GTG CTG GAC TCC GAC GGC TCC TTC TTC CTC TAC AGC AAG CTC ACC GTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   P   V   L   D   S   D   G   S   F   F   L   Y   S   K   L   T   V

       1305        1314        1323        1332        1341        1350
GAC AAG AGC AGG TGG CAG CAG GGG AAC GTC TTC TCA TGC TCC GTG ATG CAT GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   K   S   R   W   Q   Q   G   N   V   F   S   C   S   V   M   H   E
```

Fig. 8(B)

(Sheet 2 of 4)

```
      1359        1368        1377        1386        1395        1404
GCT CTG CAC AAC CAC TAC ACG CAG AAG AGC CTC TCC CTG TCT CCG AAG GGG AGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   L   H   N   H   Y   T   Q   K   S   L   S   L   S   P   K   G   S

      1413        1422        1431        1440        1449        1458
GGC GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT GGG GAG ACC AAG ATG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   G   L   K   I   A   A   F   N   I   Q   T   F   G   E   T   K   M

      1467        1476        1485        1494        1503        1512
TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG AGC CGC TAC GAC ATC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   N   A   T   L   V   S   Y   I   V   Q   I   L   S   R   Y   D   I

      1521        1530        1539        1548        1557        1566
GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC GTG GGG AAG CTG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   L   V   Q   E   V   R   D   S   H   L   T   A   V   G   K   L   L

      1575        1584        1593        1602        1611        1620
GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC GTG GTC AGT GAG CCA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   N   L   N   Q   D   A   P   D   T   Y   H   Y   V   V   S   E   P

      1629        1638        1647        1656        1665        1674
CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG TAC AGG CCT GAC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   G   R   N   S   Y   K   E   R   Y   L   F   V   Y   R   P   D   Q

      1683        1692        1701        1710        1719        1728
GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC GAG CCC TGC GGG AAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   S   A   V   D   S   Y   Y   Y   D   D   G   C   E   P   C   G   N

      1737        1746        1755        1764        1773        1782
GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC TCC CGG TTC ACA GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   T   F   N   R   E   P   A   I   V   R   F   F   S   R   F   T   E

      1791        1800        1809        1818        1827        1836
GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GCC CCG GGG GAC GCA GTA GCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   R   E   F   A   I   V   P   L   H   A   A   P   G   D   A   V   A

      1845        1854        1863        1872        1881        1890
GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA GAG AAA TGG GGC TTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   I   D   A   L   Y   D   V   Y   L   D   V   Q   E   K   W   G   L

      1899        1908        1917        1926        1935        1944
GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC AGC TAT GTG AGA CCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   D   V   M   L   M   G   D   F   N   A   G   C   S   Y   V   R   P

      1953        1962        1971        1980        1989        1998
TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC TTC CAG TGG CTG ATC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   Q   W   S   S   I   R   L   W   T   S   P   T   F   Q   W   L   I

      2007        2016        2025        2034        2043        2052
CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT GCC TAT GAC AGG ATC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   D   S   A   D   T   T   A   T   P   T   H   C   A   Y   D   R   I
```

Fig. 8(B)

(Sheet 3 of 4)

31/113

```
       2061        2070        2079        2088        2097        2106
GTG GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT GTT CCC GAC TCG GCT CTT CCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   V   A   G   M   L   L   R   G   A   V   V   P   D   S   A   L   P

       2115        2124        2133        2142        2151        2160
TTT AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC CAA CTG GCC CAA GCC ATC AGT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 F   N   F   Q   A   A   Y   G   L   S   D   Q   L   A   Q   A   I   S

       2169        2178        2187
GAC CAC TAT CCA GTG GAG GTG ATG CTG AAG TGA 3'
--- --- --- --- --- --- --- --- --- --- ---
 D   H   Y   P   V   E   V   M   L   K   *
```

Fig. 8(B)

(Sheet 4 of 4)

32/113

pAS36

```
LOCUS       PAS36.DNA    2190 bp  2190 bp    DNA                    14-AUG-
1998
DEFINITION  HUMANISED HMFG1 heavy chain fused to human DNAse - construct 36
DEFINITION  Clone 18.24.1 with residue 1392 T > C
REFERENCE
  AUTHORS   VERHOEYEN ET AL
  TITLE     CONSTRUCTION OF RESHAPED HMFG1 etc
  JOURNAL   IMMUNOL. (1993):78, 364-370
COMMENT     Human DNAse sequence is modified as a result of oligo assembly
(mhdnase.dna)
COMMENT     The fusion was made using overlapping oligos AS83 and AS84
FEATURES    AA RESIDUE 235 HAS NOT BEEN CHANGED TO KABAT (I.E. V TO A)
FEATURES    Residue 963 is G > T leading to silent mutation in all clones
FEATURES    Residue 1392 T > C silent S to S mutation
  SITES     Note
BASE COUNT      498 a    678 c    605 g    409 t
ORIGIN           ?

        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCCC ACCGTGCCCA GCACCTGAAC TCCTGGGGGG ACCGTCAGTC
      781 TTCCTCTTCC CCCCAAAACC CAAGGACACC CTCATGATCT CCCGGACCCC TGAGGTCACA
      841 TGCGTGGTGG TGGACGTGAG CCACGAAGAC CCTGAGGTCA AGTTCAACTG GTACGTGGAC
      901 GGCGTGGAGG TGCATAATGC CAAGACAAAG CCGCGGGAGG AGCAGTACAA CAGCACGTAC
      961 CGTGTGGTCA GCGTCCTCAC CGTCCTGCAC CAGGACTGGC TGAATGGCAA GGAGTACAAG
     1021 TGCAAGGTCT CCAACAAAGC CCTCCCAGCC CCCATCGAGA AAACCATCTC CAAAGCCAAA
     1081 GGGCAGCCCC GAGAACCACA GGTGTACACC CTGCCCCCAT CCCGGGATGA GCTGACCAAG
     1141 AACCAGGTCA GCCTGACCTG CCTGGTCAAA GGCTTCTATC CCAGCGACAT CGCCGTGGAG
     1201 TGGGAGAGCA ATGGGCAGCC GGAGAACAAC TACAAGACCA CGCCTCCCGT GCTGGACTCC
     1261 GACGGCTCCT TCTTCCTCTA CAGCAAGCTC ACCGTGGACA AGAGCAGGTG GCAGCAGGGG
     1321 AACGTCTTCT CATGCTCCGT GATGCATGAG GCTCTGCACA ACCACTACAC GCAGAAGAGC
     1381 CTCTCCCTGT CcCCGGGGAG CGGCGGGCTG AAGATCGCAG CCTTCAACAT CCAGACATTT
     1441 GGGGAGACCA AGATGTCCAA TGCCACCCTC GTCAGCTACA TTGTGCAGAT CCTGAGCCGC
     1501 TACGACATCG CCCTGGTCCA GGAGGTCAGA GACAGCCACC TGACTGCCGT GGGGAAGCTG
     1561 CTGGACAACC TCAATCAGGA CGCACCAGAC ACCTATCACT ACGTGGTCAG TGAGCCACTG
     1621 GGACGGAACA GCTATAAGGA GCGCTACCTG TTCGTGTACA GGCCTGACCA GGTGTCTGCG
     1681 GTGGACAGCT ACTACTACGA TGATGGCTGC GAGCCCTGCG GGAACGACAC CTTCAACCGA
     1741 GAGCCAGCCA TTGTCAGGTT CTTCTCCCGG TTCACAGAGG TCAGGGAGTT TGCCATTGTT
     1801 CCCCTGCATG CGGCCCCGGG GGACGCAGTA GCCGAGATCG ACGCTCTCTA TGACGTCTAC
     1861 CTGGATGTCC AAGAGAAATG GGGCTTGGAG GACGTCATGT TGATGGGCGA CTTCAATGCG
     1921 GGCTGCAGCT ATGTGAGACC CTCCCAGTGG TCATCCATCC GCCTGTGGAC AAGCCCCACC
     1981 TTCCAGTGGC TGATCCCCGA CAGCGCTGAC ACCACAGCTA CACCCACGCA CTGTGCCTAT
     2041 GACAGGATCG TGGTTGCAGG GATGCTGCTC CGAGGGGCCG TTGTTCCCGA CTCGGCTCTT
     2101 CCCTTTAACT TCCAGGCTGC CTATGGCCTG AGTGACCAAC TGGCCCAAGC CATCAGTGAC
     2161 CACTATCCAG TGGAGGTGAT GCTGAAGTGA
//
```

Fig. 9(A)

```
                9          18          27          36          45          54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63          72          81          90          99         108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117         126         135         144         153         162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171         180         189         198         207         216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225         234         243         252         261         270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279         288         297         306         315         324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333         342         351         360         369         378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387         396         405         414         423         432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441         450         459         468         477         486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495         504         513         522         531         540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549         558         567         576         585         594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G

              603         612         621         630         639         648
    CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

              657         666         675         684         693         702
```

Fig. 9(B)
(Sheet 1 of 4)

```
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC CCA CCG TGC CCA GCA CCT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   P   P   C   P   A   P

        765         774         783         792         801         810
GAA CTC CTG GGG GGA CCG TCA GTC TTC CTC TTC CCC CCA AAA CCC AAG GAC ACC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   L   L   G   G   P   S   V   F   L   F   P   P   K   P   K   D   T

        819         828         837         846         855         864
CTC ATG ATC TCC CGG ACC CCT GAG GTC ACA TGC GTG GTG GTG GAC GTG AGC CAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   M   I   S   R   T   P   E   V   T   C   V   V   V   D   V   S   H

        873         882         891         900         909         918
GAA GAC CCT GAG GTC AAG TTC AAC TGG TAC GTG GAC GGC GTG GAG GTG CAT AAT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   D   P   E   V   K   F   N   W   Y   V   D   G   V   E   V   H   N

        927         936         945         954         963         972
GCC AAG ACA AAG CCG CGG GAG GAG CAG TAC AAC AGC ACG TAC CGT GTG GTC AGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   K   T   K   P   R   E   E   Q   Y   N   S   T   Y   R   V   V   S

        981         990         999        1008        1017        1026
GTC CTC ACC GTC CTG CAC CAG GAC TGG CTG AAT GGC AAG GAG TAC AAG TGC AAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   L   T   V   L   H   Q   D   W   L   N   G   K   E   Y   K   C   K

       1035        1044        1053        1062        1071        1080
GTC TCC AAC AAA GCC CTC CCA GCC CCC ATC GAG AAA ACC ATC TCC AAA GCC AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   S   N   K   A   L   P   A   P   I   E   K   T   I   S   K   A   K

       1089        1098        1107        1116        1125        1134
GGG CAG CCC CGA GAA CCA CAG GTG TAC ACC CTG CCC CCA TCC CGG GAT GAG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   Q   P   R   E   P   Q   V   Y   T   L   P   P   S   R   D   E   L

       1143        1152        1161        1170        1179        1188
ACC AAG AAC CAG GTC AGC CTG ACC TGC CTG GTC AAA GGC TTC TAT CCC AGC GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   K   N   Q   V   S   L   T   C   L   V   K   G   F   Y   P   S   D

       1197        1206        1215        1224        1233        1242
ATC GCC GTG GAG TGG GAG AGC AAT GGG CAG CCG GAG AAC AAC TAC AAG ACC ACG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   A   V   E   W   E   S   N   G   Q   P   E   N   N   Y   K   T   T

       1251        1260        1269        1278        1287        1296
CCT CCC GTG CTG GAC TCC GAC GGC TCC TTC TTC CTC TAC AGC AAG CTC ACC GTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   P   V   L   D   S   D   G   S   F   F   L   Y   S   K   L   T   V

       1305        1314        1323        1332        1341        1350
GAC AAG AGC AGG TGG CAG CAG GGG AAC GTC TTC TCA TGC TCC GTG ATG CAT GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   K   S   R   W   Q   Q   G   N   V   F   S   C   S   V   M   H   E
```

Fig. 9(B)

(Sheet 2 of 4)

35/113

```
      1359        1368        1377        1386        1395        1404
GCT CTG CAC AAC CAC TAC ACG CAG AAG AGC CTC TCC CTG TCC CCG GGG AGC GGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   L   H   N   H   Y   T   Q   K   S   L   S   L   S   P   G   S   G

      1413        1422        1431        1440        1449        1458
GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT GGG GAG ACC AAG ATG TCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   L   K   I   A   A   F   N   I   Q   T   F   G   E   T   K   M   S

      1467        1476        1485        1494        1503        1512
AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG AGC CGC TAC GAC ATC GCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 N   A   T   L   V   S   Y   I   V   Q   I   L   S   R   Y   D   I   A

      1521        1530        1539        1548        1557        1566
CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC GTG GGG AAG CTG CTG GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   V   Q   E   V   R   D   S   H   L   T   A   V   G   K   L   L   D

      1575        1584        1593        1602        1611        1620
AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC GTG GTC AGT GAG CCA CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 N   L   N   Q   D   A   P   D   T   Y   H   Y   V   V   S   E   P   L

      1629        1638        1647        1656        1665        1674
GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG TAC AGG CCT GAC CAG GTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   R   N   S   Y   K   E   R   Y   L   F   V   Y   R   P   D   Q   V

      1683        1692        1701        1710        1719        1728
TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC GAG CCC TGC GGG AAC GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   A   V   D   S   Y   Y   Y   D   D   G   C   E   P   C   G   N   D

      1737        1746        1755        1764        1773        1782
ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC TCC CGG TTC ACA GAG GTC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   F   N   R   E   P   A   I   V   R   F   F   S   R   F   T   E   V

      1791        1800        1809        1818        1827        1836
AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GCC CCG GGG GAC GCA GTA GCC GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 R   E   F   A   I   V   P   L   H   A   A   P   G   D   A   V   A   E

      1845        1854        1863        1872        1881        1890
ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA GAG AAA TGG GGC TTG GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   D   A   L   Y   D   V   Y   L   D   V   Q   E   K   W   G   L   E

      1899        1908        1917        1926        1935        1944
GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC AGC TAT GTG AGA CCC TCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   V   M   L   M   G   D   F   N   A   G   C   S   Y   V   R   P   S

      1953        1962        1971        1980        1989        1998
CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC TTC CAG TGG CTG ATC CCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Q   W   S   S   I   R   L   W   T   S   P   T   F   Q   W   L   I   P

      2007        2016        2025        2034        2043        2052
GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT GCC TAT GAC AGG ATC GTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   S   A   D   T   T   A   T   P   T   H   C   A   Y   D   R   I   V
```

Fig. 9(B)

(Sheet 3 of 4)

```
      2061        2070        2079        2088        2097        2106
GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT GTT CCC GAC TCG GCT CTT CCC TTT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   A   G   M   L   L   R   G   A   V   V   P   D   S   A   L   P   F

      2115        2124        2133        2142        2151        2160
AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC CAA CTG GCC CAA GCC ATC AGT GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 N   F   Q   A   A   Y   G   L   S   D   Q   L   A   Q   A   I   S   D

      2169        2178        2187
CAC TAT CCA GTG GAG GTG ATG CTG AAG TGA 3'
--- --- --- --- --- --- --- --- --- ---
 H   Y   P   V   E   V   M   L   K   *
```

Fig. 9(B)

(Sheet 4 of 4)

37/113

pAS37

```
LOCUS       PAS37.DNA    2226 bp        2196 bp  2196 bp DNA      14-AUG-
1998
DEFINITION  HUMANISED HMFG1 heavy chain fused to human DNAse construct 37
DEFINITION  Clone 16.4.2 (same as hcdnase1.dna template file) plus NLS
REFERENCE
  AUTHORS   VERHOEYEN ET AL
  TITLE     CONSTRUCTION OF RESHAPED HMFG1 etc
  JOURNAL   IMMUNOL. (1993):78, 364-370
COMMENT     Human DNAse sequence is modified as a result of oligo assembly
(mhdnase.dna)
COMMENT     The fusion was made using overlapping oligos AS79 and AS80
FEATURES    AA RESIDUE 235 HAS NOT BEEN CHANGED TO KABAT (I.E. V TO A)
FEATURES    Residue 963 is G > T leading to silent mutation in all clones
  SITES     Note
BASE COUNT      511 a    683 c    619 g    413 t
ORIGIN          ?

       1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
      61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
     121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
     181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
     241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
     301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
     361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
     421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
     481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
     541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
     601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
     661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
     721 GACAAAACTC ACACATGCCC ACCGTGCCCA GCACCTGAAC TCCTGGGGGG ACCGTCAGTC
     781 TTCCTCTTCC CCCCAAAACC CAAGGACACC CTCATGATCT CCCGGACCCC TGAGGTCACA
     841 TGCGTGGTGG TGGACGTGAG CCACGAAGAC CCTGAGGTCA AGTTCAACTG GTACGTGGAC
     901 GGCGTGGAGG TGCATAATGC CAAGACAAAG CCGCGGGAGG AGCAGTACAA CAGCACGTAC
     961 CGTGTGGTCA GCGTCCTCAC CGTCCTGCAC CAGGACTGGC TGAATGGCAA GGAGTACAAG
    1021 TGCAAGGTCT CCAACAAAGC CCTCCCAGCC CCCATCGAGA AAACCATCTC CAAAGCCAAA
    1081 GGGCAGCCCC GAGAACCACA GGTGTACACC CTGCCCCCAT CCCGGGATGA GCTGACCAAG
    1141 AACCAGGTCA GCCTGACCTG CCTGGTCAAA GGCTTCTATC CCAGCGACAT CGCCGTGGAG
    1201 TGGGAGAGCA ATGGGCAGCC GGAGAACAAC TACAAGACCA CGCCTCCCGT GCTGGACTCC
    1261 GACGGCTCCT TCTTCCTCTA CAGCAAGCTC ACCGTGGACA AGAGCAGGTG GCAGCAGGGG
    1321 AACGTCTTCT CATGCTCCGT GATGCATGAG GCTCTGCACA ACCACTACAC GCAGAAGAGC
    1381 CTCTCCCTGT CTCCGGGTAA AGGGAGCGGC GGGCTGAAGA TCGCAGCCTT CAACATCCAG
    1441 ACATTTGGGG AGACCAAGAT GTCCAATGCC ACCCTCGTCA GCTACATTGT GCAGATCCTG
    1501 AGCCGCTACG ACATCGCCCT GGTCCAGGAG GTCAGAGACA GCCACCTGAC TGCCGTGGGG
    1561 AAGCTGCTGG ACAACCTCAA TCAGGACGCA CCAGACACCT ATCACTACGT GGTCAGTGAG
    1621 CCACTGGGAC GGAACAGCTA TAAGGAGCGC TACCTGTTCG TGTACAGGCC TGACCAGGTG
    1681 TCTGCGGTGG ACAGCTACTA CTACGATGAT GGCTGCGAGC CCTGCGGGAA CGACACCTTC
    1741 AACCGAGAGC CAGCCATTGT CAGGTTCTTC TCCCGGTTCA CAGAGGTCAG GGAGTTTGCC
    1801 ATTGTTCCCC TGCATGCGGC CCCGGGGGAC GCAGTAGCCG AGATCGACGC TCTCTATGAC
    1861 GTCTACCTGG ATGTCCAAGA GAAATGGGGC TTGGAGGACG TCATGTTGAT GGGCGACTTC
    1921 AATGCGGGCT GCAGCTATGT GAGACCCTCC CAGTGGTCAT CCATCCGCCT GTGGACAAGC
    1981 CCCACCTTCC AGTGGCTGAT CCCCGACAGC GCTGACACCA CAGCTACACC CACGCACTGT
    2041 GCCTATGACA GGATCGTGGT TGCAGGGATG CTGCTCCGAG GGGCCGTTGT TCCCGACTCG
    2101 GCTCTTCCCT TTAACTTCCA GGCTGCCTAT GGCCTGAGTG ACCAACTGGC CCAAGCCATC
    2161 AGTGACCACT ATCCAGTGGA GGTGATGCTG AAGGGGGGCG GACCCAAAAA GAAGCGCAAG
    2221 GTTTGA
//
```

NLS

Fig. 10(A)

38/113

```
                9          18         27         36         45         54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63         72         81         90         99        108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117        126        135        144        153        162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171        180        189        198        207        216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225        234        243        252        261        270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279        288        297        306        315        324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333        342        351        360        369        378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387        396        405        414        423        432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441        450        459        468        477        486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495        504        513        522        531        540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549        558        567        576        585        594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G

              603        612        621        630        639        648
    CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

              657        666        675        684        693        702
```

Fig. 10(B)
(Sheet 1 of 4)

39/113

```
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC CCA CCG TGC CCA GCA CCT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   P   P   C   P   A   P

        765         774         783         792         801         810
GAA CTC CTG GGG GGA CCG TCA GTC TTC CTC TTC CCC CCA AAA CCC AAG GAC ACC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   L   L   G   G   P   S   V   F   L   F   P   P   K   P   K   D   T

        819         828         837         846         855         864
CTC ATG ATC TCC CGG ACC CCT GAG GTC ACA TGC GTG GTG GTG GAC GTG AGC CAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   M   I   S   R   T   P   E   V   T   C   V   V   V   D   V   S   H

        873         882         891         900         909         918
GAA GAC CCT GAG GTC AAG TTC AAC TGG TAC GTG GAC GGC GTG GAG GTG CAT AAT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   D   P   E   V   K   F   N   W   Y   V   D   G   V   E   V   H   N

        927         936         945         954         963         972
GCC AAG ACA AAG CCG CGG GAG GAG CAG TAC AAC AGC ACG TAC CGT GTG GTC AGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   K   T   K   P   R   E   E   Q   Y   N   S   T   Y   R   V   V   S

        981         990         999        1008        1017        1026
GTC CTC ACC GTC CTG CAC CAG GAC TGG CTG AAT GGC AAG GAG TAC AAG TGC AAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   L   T   V   L   H   Q   D   W   L   N   G   K   E   Y   K   C   K

       1035        1044        1053        1062        1071        1080
GTC TCC AAC AAA GCC CTC CCA GCC CCC ATC GAG AAA ACC ATC TCC AAA GCC AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   S   N   K   A   L   P   A   P   I   E   K   T   I   S   K   A   K

       1089        1098        1107        1116        1125        1134
GGG CAG CCC CGA GAA CCA CAG GTG TAC ACC CTG CCC CCA TCC CGG GAT GAG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   Q   P   R   E   P   Q   V   Y   T   L   P   P   S   R   D   E   L

       1143        1152        1161        1170        1179        1188
ACC AAG AAC CAG GTC AGC CTG ACC TGC CTG GTC AAA GGC TTC TAT CCC AGC GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   K   N   Q   V   S   L   T   C   L   V   K   G   F   Y   P   S   D

       1197        1206        1215        1224        1233        1242
ATC GCC GTG GAG TGG GAG AGC AAT GGG CAG CCG GAG AAC AAC TAC AAG ACC ACG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   A   V   E   W   E   S   N   G   Q   P   E   N   N   Y   K   T   T

       1251        1260        1269        1278        1287        1296
CCT CCC GTG CTG GAC TCC GAC GGC TCC TTC TTC CTC TAC AGC AAG CTC ACC GTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   P   V   L   D   S   D   G   S   F   F   L   Y   S   K   L   T   V

       1305        1314        1323        1332        1341        1350
GAC AAG AGC AGG TGG CAG CAG GGG AAC GTC TTC TCA TGC TCC GTG ATG CAT GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   K   S   R   W   Q   Q   G   N   V   F   S   C   S   V   M   H   E
```

Fig. 10(B)

(Sheet 2 of 4)

40/113

```
      1359       1368       1377       1386       1395       1404
GCT CTG CAC AAC CAC TAC ACG CAG AAG AGC CTC TCC CTG TCT CCG GGT AAA GGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   L   H   N   H   Y   T   Q   K   S   L   S   L   S   P   G   K   G

      1413       1422       1431       1440       1449       1458
AGC GGC GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT GGG GAG ACC AAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   G   G   L   K   I   A   A   F   N   I   Q   T   F   G   E   T   K

      1467       1476       1485       1494       1503       1512
ATG TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG AGC CGC TAC GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 M   S   N   A   T   L   V   S   Y   I   V   Q   I   L   S   R   Y   D

      1521       1530       1539       1548       1557       1566
ATC GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC GTG GGG AAG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   A   L   V   Q   E   V   R   D   S   H   L   T   A   V   G   K   L

      1575       1584       1593       1602       1611       1620
CTG GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC GTG GTC AGT GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   D   N   L   N   Q   D   A   P   D   T   Y   H   Y   V   V   S   E

      1629       1638       1647       1656       1665       1674
CCA CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG TAC AGG CCT GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   L   G   R   N   S   Y   K   E   R   Y   L   F   V   Y   R   P   D

      1683       1692       1701       1710       1719       1728
CAG GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC GAG CCC TGC GGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Q   V   S   A   V   D   S   Y   Y   Y   D   D   G   C   E   P   C   G

      1737       1746       1755       1764       1773       1782
AAC GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC TCC CGG TTC ACA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 N   D   T   F   N   R   E   P   A   I   V   R   F   F   S   R   F   T

      1791       1800       1809       1818       1827       1836
GAG GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GCC CCG GGG GAC GCA GTA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   V   R   E   F   A   I   V   P   L   H   A   A   P   G   D   A   V

      1845       1854       1863       1872       1881       1890
GCC GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA GAG AAA TGG GGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   E   I   D   A   L   Y   D   V   Y   L   D   V   Q   E   K   W   G

      1899       1908       1917       1926       1935       1944
TTG GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC AGC TAT GTG AGA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   E   D   V   M   L   M   G   D   F   N   A   G   C   S   Y   V   R

      1953       1962       1971       1980       1989       1998
CCC TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC TTC CAG TGG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   S   Q   W   S   S   I   R   L   W   T   S   P   T   F   Q   W   L

      2007       2016       2025       2034       2043       2052
ATC CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT GCC TAT GAC AGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   P   D   S   A   D   T   T   A   T   P   T   H   C   A   Y   D   R
```

Fig. 10(B)

(Sheet 3 of 4)

```
       2061        2070        2079        2088        2097        2106
ATC GTG GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT GTT CCC GAC TCG GCT CTT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   V   V   A   G   M   L   L   R   G   A   V   V   P   D   S   A   L

       2115        2124        2133        2142        2151        2160
CCC TTT AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC CAA CTG GCC CAA GCC ATC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   F.  N   F   Q   A   A   Y   G   L   S   D   Q   L   A   Q   A   I

       2169        2178        2187        2196        2205        2214
AGT GAC CAC TAT CCA GTG GAG GTG ATG CTG AAG GGG GGC GGA CCC AAA AAG AAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   D   H   Y   P   V   E   V   M   L   K   G   G   G   P   K   K   K

       2223
CGC AAG GTT TGA 3'
--- --- --- ---
 R   K   V   *
```

Fig. 10(B)
(Sheet 4 of 4)

42/113

pAS38

```
LOCUS       PAS38.DNA    2223 bp   2193 bp    DNA             14-AUG-1998
DEFINITION  HUMANISED HMFG1 heavy chain fused to human DNAse construct 38
DEFINITION  Clone 17.12.1 with silent K to K mutation (1398 A > G)+NLS
REFERENCE
  AUTHORS   VERHOEYEN ET AL
  TITLE     CONSTRUCTION OF RESHAPED HMFG1 etc
  JOURNAL   IMMUNOL. (1993):78, 364-370
COMMENT     Human DNAse sequence is modified as a result of oligo assembly
(mhdnase.dna)
COMMENT     The fusion was made using overlapping oligos AS81 and AS82
FEATURES    AA RESIDUE 235 HAS NOT BEEN CHANGED TO KABAT (I.E. V TO A)
FEATURES    Residue 963 is G > T leading to silent mutation in all clones
FEATURES    In 17.12.1 residue 1398 is A > G (silent K to K mutation)
  SITES     Note
BASE COUNT      510 a    683 c    618 g    412 t
ORIGIN           ?

        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCCC ACCGTGCCCA GCACCTGAAC TCCTGGGGGG ACCGTCAGTC
      781 TTCCTCTTCC CCCCAAAACC CAAGGACACC CTCATGATCT CCCGGACCCC TGAGGTCACA
      841 TGCGTGGTGG TGGACGTGAG CCACGAAGAC CCTGAGGTCA AGTTCAACTG GTACGTGGAC
      901 GGCGTGGAGG TGCATAATGC CAAGACAAAG CCGCGGGAGG AGCAGTACAA CAGCACGTAC
      961 CGTGTGGTCA GCGTCCTCAC CGTCCTGCAC CAGGACTGGC TGAATGGCAA GGAGTACAAG
     1021 TGCAAGGTCT CCAACAAAGC CCTCCCAGCC CCCATCGAGA AAACCATCTC CAAAGCCAAA
     1081 GGGCAGCCCC GAGAACCACA GGTGTACACC CTGCCCCCAT CCCGGGATGA GCTGACCAAG
     1141 AACCAGGTCA GCCTGACCTG CCTGGTCAAA GGCTTCTATC CCAGCGACAT CGCCGTGGAG
     1201 TGGGAGAGCA ATGGGCAGCC GGAGAACAAC TACAAGACCA CGCCTCCCGT GCTGGACTCC
     1261 GACGGCTCCT TCTTCCTCTA CAGCAAGCTC ACCGTGGACA AGAGCAGGTG GCAGCAGGGG
     1321 AACGTCTTCT CATGCTCCGT GATGCATGAG GCTCTGCACA ACCACTACAC GCAGAAGAGC
     1381 CTCTCCCTGT CTCCGAAgGG GAGCGGCGGG CTGAAGATCG CAGCCTTCAA CATCCAGACA
     1441 TTTGGGGAGA CCAAGATGTC CAATGCCACC CTCGTCAGCT ACATTGTGCA GATCCTGAGC
     1501 CGCTACGACA TCGCCCTGGT CCAGGAGGTC AGAGACAGCC ACCTGACTGC CGTGGGGAAG
     1561 CTGCTGGACA ACCTCAATCA GGACGCACCA GACACCTATC ACTACGTGGT CAGTGAGCCA
     1621 CTGGGACGGA ACAGCTATAA GGAGCGCTAC CTGTTCGTGT ACAGGCCTGA CCAGGTGTCT
     1681 GCGGTGGACA GCTACTACTA CGATGATGGC TGCGAGCCCT GCGGGAACGA CACCTTCAAC
     1741 CGAGAGCCAG CCATTGTCAG GTTCTTCTCC CGGTTCACAG AGGTCAGGGA GTTTGCCATT
     1801 GTTCCCCTGC ATGCGGCCCC GGGGGACGCA GTAGCCGAGA TCGACGCTCT CTATGACGTC
     1861 TACCTGGATG TCCAAGAGAA ATGGGGCTTG GAGGACGTCA TGTTGATGGG CGACTTCAAT
     1921 GCGGGCTGCA GCTATGTGAG ACCCTCCCAG TGGTCATCCA TCCGCCTGTG GACAAGCCCC
     1981 ACCTTCCAGT GGCTGATCCC CGACAGCGCT GACACCACAG CTACACCCAC GCACTGTGCC
     2041 TATGACAGGA TCGTGGTTGC AGGGATGCTG CTCCGAGGGG CCGTTGTTCC CGACTCGGCT
     2101 CTTCCCTTTA ACTTCCAGGC TGCCTATGGC CTGAGTGACC AACTGGCCCA AGCCATCAGT
     2161 GACCACTATC CAGTGGAGGT GATGCTGAAG GGGGGCGGAC CCAAAAAGAA GCGCAAGGTT
     2221 TGA
//
```

NLS

Fig. 11(A)

```
                9         18        27        36        45        54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63        72        81        90        99        108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

               117       126       135       144       153       162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

               171       180       189       198       207       216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

               225       234       243       252       261       270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

               279       288       297       306       315       324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

               333       342       351       360       369       378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

               387       396       405       414       423       432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

               441       450       459       468       477       486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

               495       504       513       522       531       540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

               549       558       567       576       585       594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G

               603       612       621       630       639       648
    CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

               657       666       675       684       693       702
```

Fig. 11(B)
(Sheet 1 of 4)

```
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC CCA CCG TGC CCA GCA CCT
 V   E   P   K   S   C   D   K   T   H   T   C   P   P   C   P   A   P

        765         774         783         792         801         810
GAA CTC CTG GGG GGA CCG TCA GTC TTC CTC TTC CCC CCA AAA CCC AAG GAC ACC
 E   L   L   G   G   P   S   V   F   L   F   P   P   K   P   K   D   T

        819         828         837         846         855         864
CTC ATG ATC TCC CGG ACC CCT GAG GTC ACA TGC GTG GTG GTG GAC GTG AGC CAC
 L   M   I   S   R   T   P   E   V   T   C   V   V   V   D   V   S   H

        873         882         891         900         909         918
GAA GAC CCT GAG GTC AAG TTC AAC TGG TAC GTG GAC GGC GTG GAG GTG CAT AAT
 E   D   P   E   V   K   F   N   W   Y   V   D   G   V   E   V   H   N

        927         936         945         954         963         972
GCC AAG ACA AAG CCG CGG GAG GAG CAG TAC AAC AGC ACG TAC CGT GTG GTC AGC
 A   K   T   K   P   R   E   E   Q   Y   N   S   T   Y   R   V   V   S

        981         990         999        1008        1017        1026
GTC CTC ACC GTC CTG CAC CAG GAC TGG CTG AAT GGC AAG GAG TAC AAG TGC AAG
 V   L   T   V   L   H   Q   D   W   L   N   G   K   E   Y   K   C   K

       1035        1044        1053        1062        1071        1080
GTC TCC AAC AAA GCC CTC CCA GCC CCC ATC GAG AAA ACC ATC TCC AAA GCC AAA
 V   S   N   K   A   L   P   A   P   I   E   K   T   I   S   K   A   K

       1089        1098        1107        1116        1125        1134
GGG CAG CCC CGA GAA CCA CAG GTG TAC ACC CTG CCC CCA TCC CGG GAT GAG CTG
 G   Q   P   R   E   P   Q   V   Y   T   L   P   P   S   R   D   E   L

       1143        1152        1161        1170        1179        1188
ACC AAG AAC CAG GTC AGC CTG ACC TGC CTG GTC AAA GGC TTC TAT CCC AGC GAC
 T   K   N   Q   V   S   L   T   C   L   V   K   G   F   Y   P   S   D

       1197        1206        1215        1224        1233        1242
ATC GCC GTG GAG TGG GAG AGC AAT GGG CAG CCG GAG AAC AAC TAC AAG ACC ACG
 I   A   V   E   W   E   S   N   G   Q   P   E   N   N   Y   K   T   T

       1251        1260        1269        1278        1287        1296
CCT CCC GTG CTG GAC TCC GAC GGC TCC TTC TTC CTC TAC AGC AAG CTC ACC GTG
 P   P   V   L   D   S   D   G   S   F   F   L   Y   S   K   L   T   V

       1305        1314        1323        1332        1341        1350
GAC AAG AGC AGG TGG CAG CAG GGG AAC GTC TTC TCA TGC TCC GTG ATG CAT GAG
 D   K   S   R   W   Q   Q   G   N   V   F   S   C   S   V   M   H   E
```

Fig. 11(B)

(Sheet 2 of 4)

```
      1359        1368        1377        1386        1395        1404
GCT CTG CAC AAC CAC TAC ACG CAG AAG AGC CTC TCC CTG TCT CCG AAG GGG AGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   L   H   N   H   Y   T   Q   K   S   L   S   L   S   P   K   G   S

      1413        1422        1431        1440        1449        1458
GGC GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT GGG GAG ACC AAG ATG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   G   L   K   I   A   A   F   N   I   Q   T   F   G   E   T   K   M

      1467        1476        1485        1494        1503        1512
TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG AGC CGC TAC GAC ATC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   N   A   T   L   V   S   Y   I   V   Q   I   L   S   R   Y   D   I

      1521        1530        1539        1548        1557        1566
GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC GTG GGG AAG CTG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   L   V   Q   E   V   R   D   S   H   L   T   A   V   G   K   L   L

      1575        1584        1593        1602        1611        1620
GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC GTG GTC AGT GAG CCA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   N   L   N   Q   D   A   P   D   T   Y   H   Y   V   V   S   E   P

      1629        1638        1647        1656        1665        1674
CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG TAC AGG CCT GAC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   G   R   N   S   Y   K   E   R   Y   L   F   V   Y   R   P   D   Q

      1683        1692        1701        1710        1719        1728
GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC GAG CCC TGC GGG AAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   S   A   V   D   S   Y   Y   Y   D   D   G   C   E   P   C   G   N

      1737        1746        1755        1764        1773        1782
GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC TCC CGG TTC ACA GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   T   F   N   R   E   P   A   I   V   R   F   F   S   R   F   T   E

      1791        1800        1809        1818        1827        1836
GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GCC CCG GGG GAC GCA GTA GCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   R   E   F   A   I   V   P   L   H   A   A   P   G   D   A   V   A

      1845        1854        1863        1872        1881        1890
GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA GAG AAA TGG GGC TTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   I   D   A   L   Y   D   V   Y   L   D   V   Q   E   K   W   G   L

      1899        1908        1917        1926        1935        1944
GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC AGC TAT GTG AGA CCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   D   V   M   L   M   G   D   F   N   A   G   C   S   Y   V   R   P

      1953        1962        1971        1980        1989        1998
TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC TTC CAG TGG CTG ATC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   Q   W   S   S   I   R   L   W   T   S   P   T   F   Q   W   L   I

      2007        2016        2025        2034        2043        2052
CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT GCC TAT GAC AGG ATC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   D   S   A   D   T   T   A   T   P   T   H   C   A   Y   D   R   I
```

Fig. 11(C)

(Sheet 3 of 4)

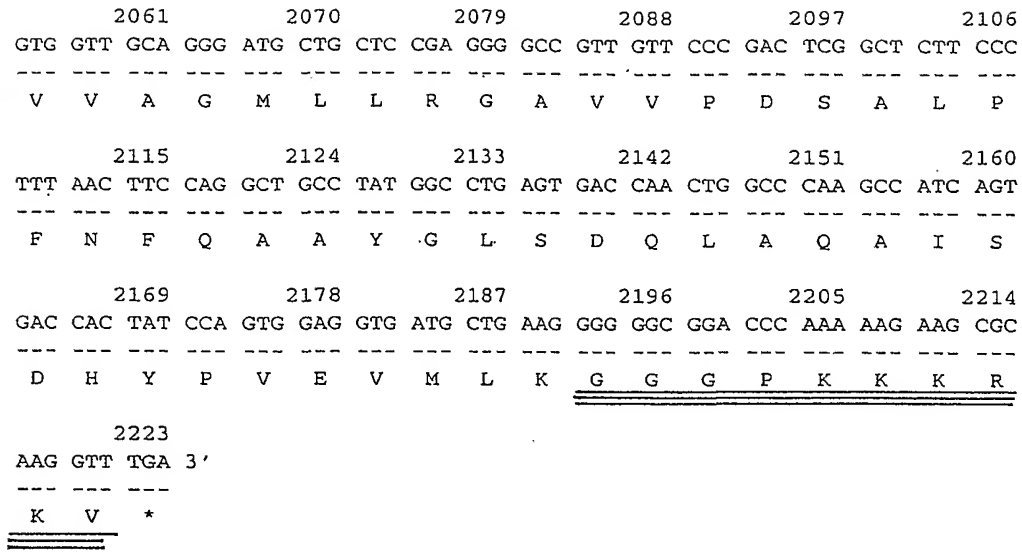

Fig. 11(D)
(Sheet 4 of 4)

47/113

pAS39

```
LOCUS       PAS39.DNA    2220 bp  2190 bp    DNA                    14-AUG-
1998
DEFINITION  HUMANISED HMFG1 heavy chain fused to human DNAse - construct 39
DEFINITION  Clone 18.24.1 with residue 1392 T > C +NLS
REFERENCE
  AUTHORS   VERHOEYEN ET AL
  TITLE     CONSTRUCTION OF RESHAPED HMFG1 etc
  JOURNAL   IMMUNOL. (1993):78, 364-370
COMMENT     Human DNAse sequence is modified as a result of oligo assembly
(mhdnase.dna)
COMMENT     The fusion was made using overlapping oligos AS83 and AS84
FEATURES    AA RESIDUE 235 HAS NOT BEEN CHANGED TO KABAT (I.E. V TO A)
FEATURES    Residue 963 is G > T leading to silent mutation in all clones
FEATURES    Residue 1392 T > C silent S to S mutation
  SITES     Note
BASE COUNT      508 a    684 c    617 g    411 t
ORIGIN          ?

        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCCC ACCGTGCCCA GCACCTGAAC TCCTGGGGGG ACCGTCAGTC
      781 TTCCTCTTCC CCCCAAAACC CAAGGACACC CTCATGATCT CCCGGACCCC TGAGGTCACA
      841 TGCGTGGTGG TGGACGTGAG CCACGAAGAC CCTGAGGTCA AGTTCAACTG GTACGTGGAC
      901 GGCGTGGAGG TGCATAATGC CAAGACAAAG CCGCGGGAGG AGCAGTACAA CAGCACGTAC
      961 CGTGTGGTCA GCGTCCTCAC CGTCCTGCAC CAGGACTGGC TGAATGGCAA GGAGTACAAG
     1021 TGCAAGGTCT CCAACAAAGC CCTCCCAGCC CCCATCGAGA AAACCATCTC CAAAGCCAAA
     1081 GGGCAGCCCC GAGAACCACA GGTGTACACC CTGCCCCCAT CCCGGGATGA GCTGACCAAG
     1141 AACCAGGTCA GCCTGACCTG CCTGGTCAAA GGCTTCTATC CCAGCGACAT CGCCGTGGAG
     1201 TGGGAGAGCA ATGGGCAGCC GGAGAACAAC TACAAGACCA CGCCTCCCGT GCTGGACTCC
     1261 GACGGCTCCT TCTTCCTCTA CAGCAAGCTC ACCGTGGACA AGAGCAGGTG GCAGCAGGGG
     1321 AACGTCTTCT CATGCTCCGT GATGCATGAG GCTCTGCACA ACCACTACAC GCAGAAGAGC
     1381 CTCTCCCTGT CcCCGGGGAG CGGCGGGCTG AAGATCGCAG CCTTCAACAT CCAGACATTT
     1441 GGGGAGACCA AGATGTCCAA TGCCACCCTC GTCAGCTACA TTGTGCAGAT CCTGAGCCGC
     1501 TACGACATCG CCCTGGTCCA GGAGGTCAGA GACAGCCACC TGACTGCCGT GGGGAAGCTG
     1561 CTGGACAACC TCAATCAGGA CGCACCAGAC ACCTATCACT ACGTGGTCAG TGAGCCACTG
     1621 GGACGGAACA GCTATAAGGA GCGCTACCTG TTCGTGTACA GGCCTGACCA GGTGTCTGCG
     1681 GTGGACAGCT ACTACTACGA TGATGGCTGC GAGCCCTGCG GGAACGACAC CTTCAACCGA
     1741 GAGCCAGCCA TTGTCAGGTT CTTCTCCCGG TTCACAGAGG TCAGGGAGTT TGCCATTGTT
     1801 CCCCTGCATG CGGCCCCGGG GGACGCAGTA GCCGAGATCG ACGCTCTCTA TGACGTCTAC
     1861 CTGGATGTCC AAGAGAAATG GGGCTTGGAG GACGTCATGT TGATGGGCGA CTTCAATGCG
     1921 GGCTGCAGCT ATGTGAGACC CTCCCAGTGG TCATCCATCC GCCTGTGGAC AAGCCCCACC
     1981 TTCCAGTGGC TGATCCCCGA CAGCGCTGAC ACCACAGCTA CACCCACGCA CTGTGCCTAT
     2041 GACAGGATCG TGGTTGCAGG GATGCTGCTC CGAGGGGCCG TTGTTCCCGA CTCGGCTCTT
     2101 CCCTTTAACT TCCAGGCTGC CTATGGCCTG AGTGACCAAC TGGCCCAAGC CATCAGTGAC
     2161 CACTATCCAG TGGAGGTGAT GCTGAAGGGG GGCGGACCCA AAAAGAAGCG CAAGGTTTGA
//
```

Fig. 12(A)

48/113

```
                9          18          27          36          45          54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63          72          81          90          99         108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117         126         135         144         153         162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171         180         189         198         207         216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225         234         243         252         261         270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279         288         297         306         315         324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333         342         351         360         369         378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387         396         405         414         423         432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441         450         459         468         477         486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495         504         513         522         531         540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549         558         567         576         585         594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G

              603         612         621         630         639         648
    CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
    L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

              657         666         675         684         693         702
```

Fig. 12(B)

(Sheet 1 of 4)

```
                9          18          27          36          45          54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63          72          81          90          99         108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117         126         135         144         153         162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171         180         189         198         207         216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225         234         243         252         261         270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279         288         297         306         315         324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333         342         351         360         369         378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387         396         405         414         423         432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441         450         459         468         477         486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495         504         513         522         531         540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549         558         567         576         585         594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G

              603         612         621         630         639         648
    CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

              657         666         675         684         693         702
```

Fig. 12(B)
(Sheet 1 of 4)

```
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC CCA CCG TGC CCA GCA CCT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   P   P   C   P   A   P

        765         774         783         792         801         810
GAA CTC CTG GGG GGA CCG TCA GTC TTC CTC TTC CCC CCA AAA CCC AAG GAC ACC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   L   L   G   G   P   S   V   F   L   F   P   P   K   P   K   D   T

        819         828         837         846         855         864
CTC ATG ATC TCC CGG ACC CCT GAG GTC ACA TGC GTG GTG GTG GAC GTG AGC CAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   M   I   S   R   T   P   E   V   T   C   V   V   V   D   V   S   H

        873         882         891         900         909         918
GAA GAC CCT GAG GTC AAG TTC AAC TGG TAC GTG GAC GGC GTG GAG GTG CAT AAT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   D   P   E   V   K   F   N   W   Y   V   D   G   V   E   V   H   N

        927         936         945         954         963         972
GCC AAG ACA AAG CCG CGG GAG GAG CAG TAC AAC AGC ACG TAC CGT GTG GTC AGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   K   T   K   P   R   E   E   Q   Y   N   S   T   Y   R   V   V   S

        981         990         999        1008        1017        1026
GTC CTC ACC GTC CTG CAC CAG GAC TGG CTG AAT GGC AAG GAG TAC AAG TGC AAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   L   T   V   L   H   Q   D   W   L   N   G   K   E   Y   K   C   K

       1035        1044        1053        1062        1071        1080
GTC TCC AAC AAA GCC CTC CCA GCC CCC ATC GAG AAA ACC ATC TCC AAA GCC AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   S   N   K   A   L   P   A   P   I   E   K   T   I   S   K   A   K

       1089        1098        1107        1116        1125        1134
GGG CAG CCC CGA GAA CCA CAG GTG TAC ACC CTG CCC CCA TCC CGG GAT GAG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   Q   P   R   E   P   Q   V   Y   T   L   P   P   S   R   D   E   L

       1143        1152        1161        1170        1179        1188
ACC AAG AAC CAG GTC AGC CTG ACC TGC CTG GTC AAA GGC TTC TAT CCC AGC GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   K   N   Q   V   S   L   T   C   L   V   K   G   F   Y   P   S   D

       1197        1206        1215        1224        1233        1242
ATC GCC GTG GAG TGG GAG AGC AAT GGG CAG CCG GAG AAC AAC TAC AAG ACC ACG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   A   V   E   W   E   S   N   G   Q   P   E   N   N   Y   K   T   T

       1251        1260        1269        1278        1287        1296
CCT CCC GTG CTG GAC TCC GAC GGC TCC TTC TTC CTC TAC AGC AAG CTC ACC GTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   P   V   L   D   S   D   G   S   F   F   L   Y   S   K   L   T   V

       1305        1314        1323        1332        1341        1350
GAC AAG AGC AGG TGG CAG CAG GGG AAC GTC TTC TCA TGC TCC GTG ATG CAT GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   K   S   R   W   Q   Q   G   N   V   F   S   C   S   V   M   H   E
```

Fig. 12(B)

(Sheet 2 of 4)

```
      1359        1368        1377        1386        1395        1404
GCT CTG CAC AAC CAC TAC ACG CAG AAG AGC CTC TCC CTG TCC CCG GGG AGC GGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   L   H   N   H   Y   T   Q   K   S   L   S   L   S   P   G   S   G

      1413        1422        1431        1440        1449        1458
GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT GGG GAG ACC AAG ATG TCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   L   K   I   A   A   F   N   I   Q   T   F   G   E   T   K   M   S

      1467        1476        1485        1494        1503        1512
AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG AGC CGC TAC GAC ATC GCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 N   A   T   L   V   S   Y   I   V   Q   I   L   S   R   Y   D   I   A

      1521        1530        1539        1548        1557        1566
CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC GTG GGG AAG CTG CTG GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   V   Q   E   V   R   D   S   H   L   T   A   V   G   K   L   L   D

      1575        1584        1593        1602        1611        1620
AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC GTG GTC AGT GAG CCA CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 N   L   N   Q   D   A   P   D   T   Y   H   Y   V   V   S   E   P   L

      1629        1638        1647        1656        1665        1674
GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG TAC AGG CCT GAC CAG GTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   R   N   S   Y   K   E   R   Y   L   F   V   Y   R   P   D   Q   V

      1683        1692        1701        1710        1719        1728
TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC GAG CCC TGC GGG AAC GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   A   V   D   S   Y   Y   Y   D   D   G   C   E   P   C   G   N   D

      1737        1746        1755        1764        1773        1782
ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC TCC CGG TTC ACA GAG GTC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   F   N   R   E   P   A   I   V   R   F   F   S   R   F   T   E   V

      1791        1800        1809        1818        1827        1836
AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GCC CCG GGG GAC GCA GTA GCC GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 R   E   F   A   I   V   P   L   H   A   A   P   G   D   A   V   A   E

      1845        1854        1863        1872        1881        1890
ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA GAG AAA TGG GGC TTG GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   D   A   L   Y   D   V   Y   L   D   V   Q   E   K   W   G   L   E

      1899        1908        1917        1926        1935        1944
GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC AGC TAT GTG AGA CCC TCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   V   M   L   M   G   D   F   N   A   G   C   S   Y   V   R   P   S

      1953        1962        1971        1980        1989        1998
CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC TTC CAG TGG CTG ATC CCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Q   W   S   S   I   R   L   W   T   S   P   T   F   Q   W   L   I   P

      2007        2016        2025        2034        2043        2052
GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT GCC TAT GAC AGG ATC GTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   S   A   D   T   T   A   T   P   T   H   C   A   Y   D   R   I   V
```

Fig. 12(B)

(Sheet 3 of 4)

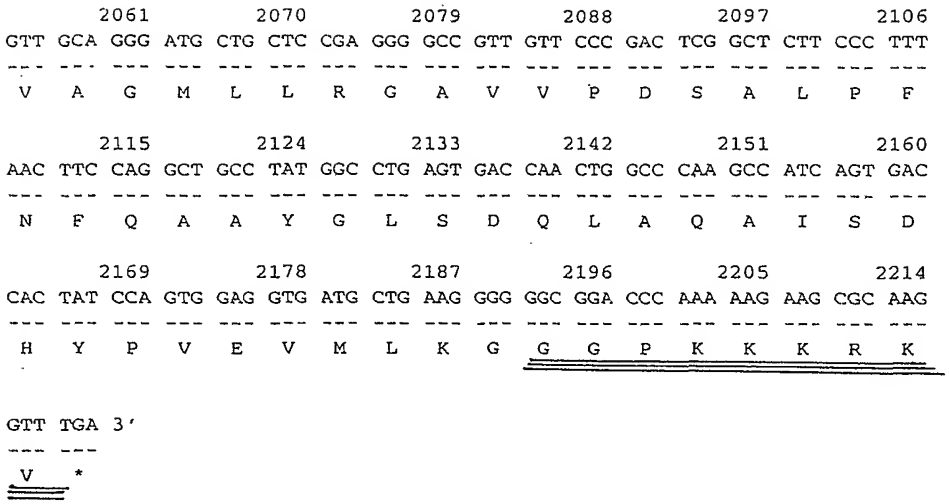

Fig. 12(B)
(Sheet 4 of 4)

52/113

pAS101

LOCUS PAS101.DNA 1548 bp mRNA PRI 06-MAR-1995
DEFINITION Humanised HMFG1 Fab'2 fused to human DNase I (pAS101)
ACCESSION
NID
KEYWORDS DNase I.
SOURCE DNase I sequence is from assembled oligos (thus modified c/f MHDNASE1.dna)
ORGANISM Homo sapiens
Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata; Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
AUTHORS Shak,S., Capon,D.J., Hellmiss,R., Marsters,S.A. and Baker,C.L.
TITLE Recombinant human DNase I reduces the viscosity of cystic fibrosis sputum
JOURNAL Proc. Natl. Acad. Sci. U.S.A. 87 (23), 9188-9192 (1990)
MEDLINE 91067672
BASE COUNT 343 a 467 c 430 g 308 t
ORIGIN

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        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCCC ACCGTGCCCA GCACCTGAAG GCGGGCTGAA GATCGCAGCC
      781 TTCAACATCC AGACATTTGG GGAGACCAAG ATGTCCAATG CCACCCTCGT CAGCTACATT
      841 GTGCAGATCC TGAGCCGCTA CGACATCGCC CTGGTCCAGG AGGTCAGAGA CAGCCACCTG
      901 ACTGCCGTGG GGAAGCTGCT GGACAACCTC AATCAGGACG CACCAGACAC CTATCACTAC
      961 GTGGTCAGTG AGCCACTGGG ACGGAACAGC TATAAGGAGC GCTACCTGTT CGTGTACAGG
     1021 CCTGACCAGG TGTCTGCGGT GGACAGCTAC TACTACGATG ATGGCTGCGA GCCCTGCGGG
     1081 AACGACACCT TCAACCGAGA GCCAGCCATT GTCAGGTTCT TCTCCCGGTT CACAGAGGTC
     1141 AGGGAGTTTG CCATTGTTCC CCTGCATGCG GCCCCGGGGG ACGCAGTAGC CGAGATCGAC
     1201 GCTCTCTATG ACGTCTACCT GGATGTCCAA GAGAAATGGG GCTTGGAGGA CGTCATGTTG
     1261 ATGGGCGACT TCAATGCGGG CTGCAGCTAT GTGAGACCCT CCCAGTGGTC ATCCATCCGC
     1321 CTGTGGACAA GCCCCACCTT CCAGTGGCTG ATCCCCGACA GCGCTGACAC CACAGCTACA
     1381 CCCACGCACT GTGCCTATGA CAGGATCGTG GTTGCAGGGA TGCTGCTCCG AGGGGCCGTT
     1441 GTTCCCGACT CGGCTCTTCC CTTTAACTTC CAGGCTGCCT ATGGCCTGAG TGACCAACTG
     1501 GCCCAAGCCA TCAGTGACCA CTATCCAGTG GAGGTGATGC TGAAGTGA
//
```

Fig. 13(A)

```
LOCUS       FDDNASE101   1548 BP SS-DNA             SYN       25-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
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     frag            721..780
                     /note="1 to 60 of 101/105linker"
     frag            join(721..>735,<736..>759,<760..>780)
                     /note="1 to 80 of 102linker [Split]"
BASE COUNT      343 A    465 C    431 G    309 T      0 OTHER
ORIGIN      -
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       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGTCC ACCGTGTCCA GCACCAGAGG GCGGGCTGAA GATCGCAGCC
      781 TTCAACATCC AGACATTTGG GGAGACCAAG ATGTCCAATG CCACCCTCGT CAGCTACATT
      841 GTGCAGATCC TGAGCCGCTA CGACATCGCC CTGGTCCAGG AGGTCAGAGA CAGCCACCTG
      901 ACTGCCGTGG GGAAGCTGCT GGACAACCTC AATCAGGACG CACCAGACAC CTATCACTAC
      961 GTGGTCAGTG AGCCACTGGG ACGGAACAGC TATAAGGAGC GCTACCTGTT CGTGTACAGG
     1021 CCTGACCAGG TGTCTGCGGT GGACAGCTAC TACTACGATG ATGGCTGCGA GCCCTGCGGG
     1081 AACGACACCT TCAACCGAGA GCCAGCCATT GTCAGGTTCT TCTCCCGGTT CACAGAGGTC
     1141 AGGGAGTTTG CCATTGTTCC CCTGCATGCG GCCCCGGGGG ACGCAGTAGC CGAGATCGAC
     1201 GCTCTCTATG ACGTCTACCT GGATGTCCAA GAGAAATGGG GCTTGGAGGA CGTCATGTTG
     1261 ATGGGCGACT TCAATGCGGG CTGCAGCTAT GTGAGACCCT CCCAGTGGTC ATCCATCCGC
     1321 CTGTGGACAA GCCCCACCTT CCAGTGGCTG ATCCCCGACA GCGCTGACAC CACAGCTACA
     1381 CCCACGCACT GTGCCTATGA CAGGATCGTG GTTGCAGGGA TGCTGCTCCG AGGGGCCGTT
     1441 GTTCCCGACT CGGCTCTTCC CTTTAACTTC CAGGCTGCCT ATGGCCTGAG TGACCAACTG
     1501 GCCCAAGCCA TCAGTGACCA CTATCCAGTG GAGGTGATGC TGAAGTGA
//
```

Fig. 13(B)

54/113

```
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DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
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     frag            join(10..>729,<790..1557)
                     /note="1 to 1548 of PAS101.dna [Split]"
     frag            730..789
                     /note="1 to 60 of 101/105linker"
     frag            join(730..>744,<745..>768,<769..>789)
                     /note="1 to 80 of 102linker [Split]"
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ORIGIN      -
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       61 CACTCCCAGG TGCAGCTGGT GCAGTCTGGG GCAGAGGTGA AAAAGCCTGG GGCCTCAGTG
      121 AAGGTGTCCT GCAAGGCTTC TGGCTACACC TTCAGTGCCT ACTGGATAGA GTGGGTGCGC
      181 CAGGCTCCAG GAAAGGGCCT CGAGTGGGTC GGAGAGATTT TACCTGGAAG TAATAATTCT
      241 AGATACAATG AGAAGTTCAA GGGCCGAGTG ACAGTCACTA GAGACACATC CACAAACACA
      301 GCCTACATGG AGCTCAGCAG CCTGAGGTCT GAGGACACAG CCGTCTATTA CTGTGCAAGA
      361 TCCTACGACT TTGCCTGGTT TGCTTACTGG GGCCAAGGGA CTCTGGTCAC AGTCTCCTCA
      421 GCCTCCACCA AGGGCCCATC GGTCTTCCCC CTGGCACCCT CCTCCAAGAG CACCTCTGGG
      481 GGCACAGCGG CCCTGGGCTG CCTGGTCAAG GACTACTTCC CCGAACCGGT GACGGTGTCG
      541 TGGAACTCAG GCGCCCTGAC CAGCGGCGTG CACACCTTCC CGGCTGTCCT ACAGTCCTCA
      601 GGACTCTACT CCCTCAGCAG CGTGGTGACC GTGCCCTCCA GCAGCTTGGG CACCCAGACC
      661 TACATCTGCA ACGTGAATCA CAAGCCCAGC AACACCAAGG TGGACAAGAA AGTTGAGCCC
      721 AAATCTTGTG ACAAAACTCA CACATGTCCA CCGTGTCCAG CACCAGAGGG CGGGCTGAAG
      781 ATCGCAGCCT TCAACATCCA GACATTTGGG GAGACCAAGA TGTCCAATGC CACCCTCGTC
      841 AGCTACATTG TGCAGATCCT GAGCCGCTAC GACATCGCCC TGGTCCAGGA GGTCAGAGAC
      901 AGCCACCTGA CTGCCGTGGG GAAGCTGCTG GACAACCTCA ATCAGGACGC ACCAGACACC
      961 TATCACTACG TGGTCAGTGA GCCACTGGGA CGGAACAGCT ATAAGGAGCG CTACCTGTTC
     1021 GTGTACAGGC CTGACCAGGT GTCTGCGGTG GACAGCTACT ACTACGATGA TGGCTGCGAG
     1081 CCCTGCGGGA ACGACACCTT CAACCGAGAG CCAGCCATTG TCAGGTTCTT CTCCCGGTTC
     1141 ACAGAGGTCA GGGAGTTTGC CATTGTTCCC CTGCATGCGG CCCCGGGGGA CGCAGTAGCC
     1201 GAGATCGACG CTCTCTATGA CGTCTACCTG GATGTCCAAG AGAAATGGGG CTTGGAGGAC
     1261 GTCATGTTGA TGGGCGACTT CAATGCGGGC TGCAGCTATG TGAGACCCTC CCAGTGGTCA
     1321 TCCATCCGCC TGTGGACAAG CCCCACCTTC CAGTGGCTGA TCCCCGACAG CGCTGACACC
     1381 ACAGCTACAC CCACGCACTG TGCCTATGAC AGGATCGTGG TTGCAGGGAT GCTGCTCCGA
     1441 GGGGCCGTTG TTCCCGACTC GGCTCTTCCC TTTAACTTCC AGGCTGCCTA TGGCCTGAGT
     1501 GACCAACTGG CCCAAGCCAT CAGTGACCAC TATCCAGTGG AGGTGATGCT GAAGTGA
//
```

Fig. 13(C)

```
                9          18         27         36         45         54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63         72         81         90         99        108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117        126        135        144        153        162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171        180        189        198        207        216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225        234        243        252        261        270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279        288        297        306        315        324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333        342        351        360        369        378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387        396        405        414        423        432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441        450        459        468        477        486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495        504        513        522        531        540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549        558        567        576        585        594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G
```

Fig. 13(D)
(Sheet 1 of 3)

56/113

```
        603         612         621         630         639         648
CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

        657         666         675         684         693         702
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC CCA CCG TGC CCA GCA CCT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   P   P   C   P   A   P

        765         774         783         792         801         810
GAA GGC GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT GGG GAG ACC AAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   G   G   L   K   I   A   A   F   N   I   Q   T   F   G   E   T   K

        819         828         837         846         855         864
ATG TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG AGC CGC TAC GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 M   S   N   A   T   L   V   S   Y   I   V   Q   I   L   S   R   Y   D

        873         882         891         900         909         918
ATC GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC GTG GGG AAG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   A   L   V   Q   E   V   R   D   S   H   L   T   A   V   G   K   L

        927         936         945         954         963         972
CTG GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC GTG GTC AGT GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   D   N   L   N   Q   D   A   P   D   T   Y   H   Y   V   V   S   E

        981         990         999        1008        1017        1026
CCA CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG TAC AGG CCT GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   L   G   R   N   S   Y   K   E   R   Y   L   F   V   Y   R   P   D

       1035        1044        1053        1062        1071        1080
CAG GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC GAG CCC TGC GGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Q   V   S   A   V   D   S   Y   Y   Y   D   D   G   C   E   P   C   G

       1089        1098        1107        1116        1125        1134
AAC GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC TCC CGG TTC ACA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 N   D   T   F   N   R   E   P   A   I   V   R   F   F   S   R   F   T

       1143        1152        1161        1170        1179        1188
GAG GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GCC CCG GGG GAC GCA GTA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   V   R   E   F   A   I   V   P   L   H   A   A   P   G   D   A   V

       1197        1206        1215        1224        1233        1242
```

Fig. 13(D)

(Sheet 2 of 3)

```
GCC GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA GAG AAA TGG GGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   E   I   D   A   L   Y   D   V   Y   L   D   V   Q   E   K   W   G

       1251        1260        1269        1278        1287        1296
TTG GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC AGC TAT GTG AGA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   E   D   V   M   L   M   G   D   F   N   A   G   C   S   Y   V   R

       1305        1314        1323        1332        1341        1350
CCC TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC TTC CAG TGG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   S   Q   W   S   S   I   R   L   W   T   S   P   T   F   Q   W   L

       1359        1368        1377        1386        1395        1404
ATC CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT GCC TAT GAC AGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   P   D   S   A   D   T   T   A   T   P   T   H   C   A   Y   D   R

       1413        1422        1431        1440        1449        1458
ATC GTG GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT GTT CCC GAC TCG GCT CTT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   V   V   A   G   M   L   L   R   G   A   V   V   P   D   S   A   L

       1467        1476        1485        1494        1503        1512
CCC TTT AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC CAA CTG GCC CAA GCC ATC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   F   N   F   Q   A   A   Y   G   L   S   D   Q   L   A   Q   A   I

       1521        1530        1539        1548
AGT GAC CAC TAT CCA GTG GAG GTG ATG CTG AAG TGA 3'
--- --- --- --- --- --- --- --- --- --- --- ---
 S   D   H   Y   P   V   E   V   M   L   K   *
```

Fig. 13(D)
(Sheet 3 of 3)

58/113

pAS102

```
LOCUS       PAS102.DNA   1566 bp    mRNA            PRI       06-MAR-1995
DEFINITION  Humanised HMFG1 Fab'2 fused to human DNase I (pAS102)
ACCESSION
NID
KEYWORDS    DNase I.
SOURCE      DNase I sequence is from assembled oligos (thus modified c/f
MHDNASE1.dna)  (See Figure 2)
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
  AUTHORS   Shak,S., Capon,D.J., Hellmiss,R., Marsters,S.A. and Baker,C.L.
  TITLE     Recombinant human DNase I reduces the viscosity of cystic
fibrosis
            sputum
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 87 (23), 9188-9192 (1990)
  MEDLINE   91067672
BASE COUNT      345 a    469 c    440 g    312 t
ORIGIN

        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCTG TGTGGAGTGC CCACCGTGCC CAGCACCTGA AGGGAGCGGC
      781 GGGCTGAAGA TCGCAGCCTT CAACATCCAG ACATTTGGGG AGACCAAGAT GTCCAATGCC
      841 ACCCTCGTCA GCTACATTGT GCAGATCCTG AGCCGCTACG ACATCGCCCT GGTCCAGGAG
      901 GTCAGAGACA GCCACCTGAC TGCCGTGGGG AAGCTGCTGG ACAACCTCAA TCAGGACGCA
      961 CCAGACACCT ATCACTACGT GGTCAGTGAG CCACTGGGAC GGAACAGCTA TAAGGAGCGC
     1021 TACCTGTTCG TGTACAGGCC TGACCAGGTG TCTGCGGTGG ACAGCTACTA CTACGATGAT
     1081 GGCTGCGAGC CCTGCGGGAA CGACACCTTC AACCGAGAGC CAGCCATTGT CAGGTTCTTC
     1141 TCCCGGTTCA CAGAGGTCAG GGAGTTTGCC ATTGTTCCCC TGCATGCGGC CCCGGGGGAC
     1201 GCAGTAGCCG AGATCGACGC TCTCTATGAC GTCTACCTGG ATGTCCAAGA GAAATGGGGC
     1261 TTGGAGGACG TCATGTTGAT GGGCGACTTC AATGCGGGCT GCAGCTATGT GAGACCCTCC
     1321 CAGTGGTCAT CCATCCGCCT GTGGACAAGC CCCACCTTCC AGTGGCTGAT CCCCGACAGC
     1381 GCTGACACCA CAGCTACACC CACGCACTGT GCCTATGACA GGATCGTGGT TGCAGGGATG
     1441 CTGCTCCGAG GGGCCGTTGT TCCCGACTCG GCTCTTCCCT TTAACTTCCA GGCTGCCTAT
     1501 GGCCTGAGTG ACCAACTGGC CCAAGCCATC AGTGACCACT ATCCAGTGGA GGTGATGCTG
     1561 AAGTGA
//
```

Fig. 14(A)

59/113

```
LOCUS       FDDNASE102    1566 BP SS-DNA              SYN       23-MAR-2001
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
BASE COUNT       345 A    468 C    440 G    313 T      0 OTHER
ORIGIN      -
        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCTG TGTCGAGTGT CCACCGTGTC CAGCACCAGA GGGGAGCGGC
      781 GGGCTGAAGA TCGCAGCCTT CAACATCCAG ACATTTGGGG AGACCAAGAT GTCCAATGCC
      841 ACCCTCGTCA GCTACATTGT GCAGATCCTG AGCCGCTACG ACATCGCCCT GGTCCAGGAG
      901 GTCAGAGACA GCCACCTGAC TGCCGTGGGG AAGCTGCTGG ACAACCTCAA TCAGGACGCA
      961 CCAGACACCT ATCACTACGT GGTCAGTGAG CCACTGGGAC GGAACAGCTA TAAGGAGCGC
     1021 TACCTGTTCG TGTACAGGCC TGACCAGGTG TCTGCGGTGG ACAGCTACTA CTACGATGAT
     1081 GGCTGCGAGC CCTGCGGGAA CGACACCTTC AACCGAGAGC CAGCCATTGT CAGGTTCTTC
     1141 TCCCGGTTCA CAGAGGTCAG GGAGTTTGCC ATTGTTCCCC TGCATGCGGC CCCGGGGGAC
     1201 GCAGTAGCCG AGATCGACGC TCTCTATGAC GTCTACCTGG ATGTCCAAGA GAAATGGGGC
     1261 TTGGAGGACG TCATGTTGAT GGGCGACTTC AATGCGGGCT GCAGCTATGT GAGACCCTCC
     1321 CAGTGGTCAT CCATCCGCCT GTGGACAAGC CCCACCTTCC AGTGGCTGAT CCCCGACAGC
     1381 GCTGACACCA CAGCTACACC CACGCACTGT GCCTATGACA GGATCGTGGT TGCAGGGATG
     1441 CTGCTCCGAG GGGCCGTTGT TCCCGACTCG GCTCTTCCCT TTAACTTCCA GGCTGCCTAT
     1501 GGCCTGAGTG ACCAACTGGC CCAAGCCATC AGTGACCACT ATCCAGTGGA GGTGATGCTG
     1561 AAGTGA
//
```

Fig. 14(B)

60/113

pAS302

```
LOCUS       FDDNASE302   1575 BP SS-DNA             SYN       29-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
     frag            10..1575
                     /note="1 to 1566 of FdDNase102correct"
BASE COUNT      346 A    474 C    442 G    313 T      0 OTHER
ORIGIN      -
        1 GCCGCCACCA TGGGATGGAG CTGTATCATC CTCTTCTTGG TAGCAACAGC TACAGGTGTC
       61 CACTCCCAGG TGCAGCTGGT GCAGTCTGGG GCAGAGGTGA AAAAGCCTGG GGCCTCAGTG
      121 AAGGTGTCCT GCAAGGCTTC TGGCTACACC TTCAGTGCCT ACTGGATAGA GTGGGTGCGC
      181 CAGGCTCCAG GAAAGGGCCT CGAGTGGGTC GGAGAGATTT TACCTGGAAG TAATAATTCT
      241 AGATACAATG AGAAGTTCAA GGGCCGAGTG ACAGTCACTA GAGACACATC CACAAACACA
      301 GCCTACATGG AGCTCAGCAG CCTGAGGTCT GAGGACACAG CCGTCTATTA CTGTGCAAGA
      361 TCCTACGACT TTGCCTGGTT TGCTTACTGG GGCCAAGGGA CTCTGGTCAC AGTCTCCTCA
      421 GCCTCCACCA AGGGCCCATC GGTCTTCCCC CTGGCACCCT CCTCCAAGAG CACCTCTGGG
      481 GGCACAGCGG CCCTGGGCTG CCTGGTCAAG GACTACTTCC CCGAACCGGT GACGGTGTCG
      541 TGGAACTCAG GCGCCCTGAC CAGCGGCGTG CACACCTTCC CGGCTGTCCT ACAGTCCTCA
      601 GGACTCTACT CCCTCAGCAG CGTGGTGACC GTGCCCTCCA GCAGCTTGGG CACCCAGACC
      661 TACATCTGCA ACGTGAATCA CAAGCCCAGC AACACCAAGG TGGACAAGAA AGTTGAGCCC
      721 AAATCTTGTG ACAAAACTCA CACATGCTGT GTCGAGTGTC CACCGTGTCC AGCACCAGAG
      781 GGGAGCGGCG GGCTGAAGAT CGCAGCCTTC AACATCCAGA CATTTGGGGA GACCAAGATG
      841 TCCAATGCCA CCCTCGTCAG CTACATTGTG CAGATCCTGA GCCGCTACGA CATCGCCCTG
      901 GTCCAGGAGG TCAGAGACAG CCACCTGACT GCCGTGGGGA AGCTGCTGGA CAACCTCAAT
      961 CAGGACGCAC CAGACACCTA TCACTACGTG GTCAGTGAGC CACTGGGACG GAACAGCTAT
     1021 AAGGAGCGCT ACCTGTTCGT GTACAGGCCT GACCAGGTGT CTGCGGTGGA CAGCTACTAC
     1081 TACGATGATG GCTGCGAGCC CTGCGGGAAC GACACCTTCA ACCGAGAGCC AGCCATTGTC
     1141 AGGTTCTTCT CCCGGTTCAC AGAGGTCAGG GAGTTTGCCA TTGTTCCCCT GCATGCGGCC
     1201 CCGGGGGACG CAGTAGCCGA GATCGACGCT CTCTATGACG TCTACCTGGA TGTCCAAGAG
     1261 AAATGGGGCT TGGAGGACGT CATGTTGATG GGCGACTTCA ATGCGGGCTG CAGCTATGTG
     1321 AGACCCTCCC AGTGGTCATC CATCCGCCTG TGGACAAGCC CCACCTTCCA GTGGCTGATC
     1381 CCCGACAGCG CTGACACCAC AGCTACACCC ACGCACTGTG CCTATGACAG GATCGTGGTT
     1441 GCAGGGATGC TGCTCCGAGG GGCCGTTGTT CCCGACTCGG CTCTTCCCTT TAACTTCCAG
     1501 GCTGCCTATG GCCTGAGTGA CCAACTGGCC CAAGCCATCA GTGACCACTA TCCAGTGGAG
     1561 GTGATGCTGA AGTGA
//
```

Fig. 14(C)

```
                9          18          27          36          45          54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63          72          81          90          99         108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117         126         135         144         153         162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171         180         189         198         207         216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225         234         243         252         261         270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279         288         297         306         315         324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333         342         351         360         369         378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387         396         405         414         423         432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441         450         459         468         477         486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495         504         513         522         531         540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549         558         567         576         585         594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G
```

Fig. 14(D)

(Sheet 1 of 3)

62/113

```
        603         612         621         630         639         648
CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

        657         666         675         684         693         702
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC TGT GTG GAG TGC CCA CCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   C   V   E   C   P   P

        765         774         783         792         801         810
TGC CCA GCA CCT GAA GGG AGC GGC GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 C   P   A   P   E   G   S   G   G   L   K   I   A   A   F   N   I   Q

        819         828         837         846         855         864
ACA TTT GGG GAG ACC AAG ATG TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   F   G   E   T   K   M   S   N   A   T   L   V   S   Y   I   V   Q

        873         882         891         900         909         918
ATC CTG AGC CGC TAC GAC ATC GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   L   S   R   Y   D   I   A   L   V   Q   E   V   R   D   S   H   L

        927         936         945         954         963         972
ACT GCC GTG GGG AAG CTG CTG GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   A   V   G   K   L   L   D   N   L   N   Q   D   A   P   D   T   Y

        981         990         999        1008        1017        1026
CAC TAC GTG GTC AGT GAG CCA CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 H   Y   V   V   S   E   P   L   G   R   N   S   Y   K   E   R   Y   L

       1035        1044        1053        1062        1071        1080
TTC GTG TAC AGG CCT GAC CAG GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 F   V   Y   R   P   D   Q   V   S   A   V   D   S   Y   Y   Y   D   D

       1089        1098        1107        1116        1125        1134
GGC TGC GAG CCC TGC GGG AAC GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   C   E   P   C   G   N   D   T   F   N   R   E   P   A   I   V   R

       1143        1152        1161        1170        1179        1188
TTC TTC TCC CGG TTC ACA GAG GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 F   F   S   R   F   T   E   V   R   E   F   A   I   V   P   L   H   A

       1197        1206        1215        1224        1233        1242
```

Fig. 14(D)

(Sheet 2 of 3)

```
GCC CCG GGG GAC GCA GTA GCC GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   P   G   D   A   V   A   E   I   D   A   L   Y   D   V   Y   L   D

       1251        1260        1269        1278        1287        1296
GTC CAA GAG AAA TGG GGC TTG GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   Q   E   K   W   G   L   E   D   V   M   L   M   G   D   F   N   A

       1305        1314        1323        1332        1341        1350
GGC TGC AGC TAT GTG AGA CCC TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   C   S   Y   V   R   P   S   Q   W   S   S   I   R   L   W   T   S

       1359        1368        1377        1386        1395        1404
CCC ACC TTC CAG TGG CTG ATC CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   T   F   Q   W   L   I   P   D   S   A   D   T   T   A   T   P   T

       1413        1422        1431        1440        1449        1458
CAC TGT GCC TAT GAC AGG ATC GTG GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 H   C   A   Y   D   R   I   V   V   A   G   M   L   L   R   G   A   V

       1467        1476        1485        1494        1503        1512
GTT CCC GAC TCG GCT CTT CCC TTT AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   P   D   S   A   L   P   F   N   F   Q   A   A   Y   G   L   S   D

       1521        1530        1539        1548        1557        1566
CAA CTG GCC CAA GCC ATC AGT GAC CAC TAT CCA GTG GAG GTG ATG CTG AAG TGA 3'
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Q   L   A   Q   A   I   S   D   H   Y   P   V   E   V   M   L   K   *
```

Fig. 14(D)
(Sheet 3 of 3)

64/113

pAS103

```
LOCUS       PAS103.DNA   1560 bp    mRNA            PRI       06-MAR-1995
DEFINITION  Humanised HMFG1 Fab'2 fused to human DNase I (pAS103)
ACCESSION
NID
KEYWORDS    DNase I.
SOURCE      DNase I sequence is from assembled oligos (thus modified c/f
MHDNASE1.dna)
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
  AUTHORS   Shak,S., Capon,D.J., Hellmiss,R., Marsters,S.A. and Baker,C.L.
  TITLE     Recombinant human DNase I reduces the viscosity of cystic
fibrosis
            sputum
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 87 (23), 9188-9192 (1990)
  MEDLINE   91067672
BASE COUNT      344 a    468 c    436 g    312 t
ORIGIN

        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCTG TGTGGAGTGC CCACCGTGCC CAGCACCTGA AGGCGGGCTG
      781 AAGATCGCAG CCTTCAACAT CCAGACATTT GGGGAGACCA AGATGTCCAA TGCCACCCTC
      841 GTCAGCTACA TTGTGCAGAT CCTGAGCCGC TACGACATCG CCCTGGTCCA GGAGGTCAGA
      901 GACAGCCACC TGACTGCCGT GGGGAAGCTG CTGGACAACC TCAATCAGGA CGCACCAGAC
      961 ACCTATCACT ACGTGGTCAG TGAGCCACTG GGACGGAACA GCTATAAGGA GCGCTACCTG
     1021 TTCGTGTACA GGCCTGACCA GGTGTCTGCG GTGGACAGCT ACTACTACGA TGATGGCTGC
     1081 GAGCCCTGCG GGAACGACAC CTTCAACCGA GAGCCAGCCA TTGTCAGGTT CTTCTCCCGG
     1141 TTCACAGAGG TCAGGGAGTT TGCCATTGTT CCCCTGCATG CGGCCCCGGG GGACGCAGTA
     1201 GCCGAGATCG ACGCTCTCTA TGACGTCTAC CTGGATGTCC AAGAGAAATG GGGCTTGGAG
     1261 GACGTCATGT TGATGGGCGA CTTCAATGCG GGCTGCAGCT ATGTGAGACC CTCCCAGTGG
     1321 TCATCCATCC GCCTGTGGAC AAGCCCCACC TTCCAGTGGC TGATCCCCGA CAGCGCTGAC
     1381 ACCACAGCTA CACCCACGCA CTGTGCCTAT GACAGGATCG TGGTTGCAGG GATGCTGCTC
     1441 CGAGGGGCCG TTGTTCCCGA CTCGGCTCTT CCCTTTAACT TCCAGGCTGC CTATGGCCTG
     1501 AGTGACCAAC TGGCCCAAGC CATCAGTGAC CACTATCCAG TGGAGGTGAT GCTGAAGTGA
//
```

Fig. 15(A)

65/113

```
LOCUS       FDDNASE103   1560 BP SS-DNA             SYN       25-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
     frag            join(1..>720,<793..1560)
                     /note="1 to 1560 of PAS103.dna [Split]"
     frag            721..792
                     /note="1 to 72 of 103/107linker"
     frag            join(721..>771,<772..792)
                     /note="1 to 78 of 102linker [Split]"
BASE COUNT      344 A    467 C    436 G    313 T      0 OTHER
ORIGIN      -
        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCTG TGTCGAGTGT CCACCGTGTC CAGCACCAGA GGGCGGGCTG
      781 AAGATCGCAG CCTTCAACAT CCAGACATTT GGGGAGACCA AGATGTCCAA TGCCACCCTC
      841 GTCAGCTACA TTGTGCAGAT CCTGAGCCGC TACGACATCG CCCTGGTCCA GGAGGTCAGA
      901 GACAGCCACC TGACTGCCGT GGGGAAGCTG CTGGACAACC TCAATCAGGA CGCACCAGAC
      961 ACCTATCACT ACGTGGTCAG TGAGCCACTG GGACGGAACA GCTATAAGGA GCGCTACCTG
     1021 TTCGTGTACA GGCCTGACCA GGTGTCTGCG GTGGACAGCT ACTACTACGA TGATGGCTGC
     1081 GAGCCCTGCG GGAACGACAC CTTCAACCGA GAGCCAGCCA TTGTCAGGTT CTTCTCCCGG
     1141 TTCACAGAGG TCAGGGAGTT TGCCATTGTT CCCCTGCATG CGGCCCCGGG GGACGCAGTA
     1201 GCCGAGATCG ACGCTCTCTA TGACGTCTAC CTGGATGTCC AAGAGAAATG GGGCTTGGAG
     1261 GACGTCATGT TGATGGGCGA CTTCAATGCG GGCTGCAGCT ATGTGAGACC CTCCCAGTGG
     1321 TCATCCATCC GCCTGTGGAC AAGCCCCACC TTCCAGTGGC TGATCCCCGA CAGCGCTGAC
     1381 ACCACAGCTA CACCCACGCA CTGTGCCTAT GACAGGATCG TGGTTGCAGG GATGCTGCTC
     1441 CGAGGGGCCG TTGTTCCCGA CTCGGCTCTT CCCTTTAACT TCCAGGCTGC CTATGGCCTG
     1501 AGTGACCAAC TGGCCCAAGC CATCAGTGAC CACTATCCAG TGGAGGTGAT GCTGAAGTGA
//
```

Fig. 15(B)

66/113

```
LOCUS       FDDNASE103   1569 BP SS-DNA             SYN       29-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
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                     /note="1 to 1560 of FdDNase103correct"
     frag            join(10..>729,<802..1569)
                     /note="1 to 1560 of PAS103.dna [Split]"
     frag            730..801
                     /note="1 to 72 of 103/107linker"
     frag            join(730..>780,<781..801)
                     /note="1 to 78 of 102linker [Split]"
BASE COUNT      345 A    473 C    438 G    313 T      0 OTHER
ORIGIN      -
        1 GCCGCCACCA TGGGATGGAG CTGTATCATC CTCTTCTTGG TAGCAACAGC TACAGGTGTC
       61 CACTCCCAGG TGCAGCTGGT GCAGTCTGGG GCAGAGGTGA AAAAGCCTGG GGCCTCAGTG
      121 AAGGTGTCCT GCAAGGCTTC TGGCTACACC TTCAGTGCCT ACTGGATAGA GTGGGTGCGC
      181 CAGGCTCCAG GAAAGGGCCT CGAGTGGGTC GGAGAGATTT TACCTGGAAG TAATAATTCT
      241 AGATACAATG AGAAGTTCAA GGGCCGAGTG ACAGTCACTA GAGACACATC CACAAACACA
      301 GCCTACATGG AGCTCAGCAG CCTGAGGTCT GAGGACACAG CCGTCTATTA CTGTGCAAGA
      361 TCCTACGACT TTGCCTGGTT TGCTTACTGG GGCCAAGGGA CTCTGGTCAC AGTCTCCTCA
      421 GCCTCCACCA AGGGCCCATC GGTCTTCCCC CTGGCACCCT CCTCCAAGAG CACCTCTGGG
      481 GGCACAGCGG CCCTGGGCTG CCTGGTCAAG GACTACTTCC CCGAACCGGT GACGGTGTCG
      541 TGGAACTCAG GCGCCCTGAC CAGCGGCGTG CACACCTTCC CGGCTGTCCT ACAGTCCTCA
      601 GGACTCTACT CCCTCAGCAG CGTGGTGACC GTGCCCTCCA GCAGCTTGGG CACCCAGACC
      661 TACATCTGCA ACGTGAATCA CAAGCCCAGC AACACCAAGG TGGACAAGAA AGTTGAGCCC
      721 AAATCTTGTG ACAAAACTCA CACATGCTGT GTCGAGTGTC CACCGTGTCC AGCACCAGAG
      781 GGCGGGCTGA AGATCGCAGC CTTCAACATC CAGACATTTG GGGAGACCAA GATGTCCAAT
      841 GCCACCCTCG TCAGCTACAT TGTGCAGATC CTGAGCCGCT ACGACATCGC CCTGGTCCAG
      901 GAGGTCAGAG ACAGCCACCT GACTGCCGTG GGGAAGCTGC TGGACAACCT CAATCAGGAC
      961 GCACCAGACA CCTATCACTA CGTGGTCAGT GAGCCACTGG GACGGAACAG CTATAAGGAG
     1021 CGCTACCTGT TCGTGTACAG GCCTGACCAG GTGTCTGCGG TGGACAGCTA CTACTACGAT
     1081 GATGGCTGCG AGCCCTGCGG GAACGACACC TTCAACCGAG AGCCAGCCAT TGTCAGGTTC
     1141 TTCTCCCGGT TCACAGAGGT CAGGGAGTTT GCCATTGTTC CCCTGCATGC GGCCCCGGGG
     1201 GACGCAGTAG CCGAGATCGA CGCTCTCTAT GACGTCTACC TGGATGTCCA AGAGAAATGG
     1261 GGCTTGGAGG ACGTCATGTT GATGGGCGAC TTCAATGCGG GCTGCAGCTA TGTGAGACCC
     1321 TCCCAGTGGT CATCCATCCG CCTGTGGACA AGCCCCACCT TCCAGTGGCT GATCCCCGAC
     1381 AGCGCTGACA CCACAGCTAC ACCCACGCAC TGTGCCTATG ACAGGATCGT GGTTGCAGGG
     1441 ATGCTGCTCC GAGGGGCCGT TGTTCCCGAC TCGGCTCTTC CCTTTAACTT CCAGGCTGCC
     1501 TATGGCCTGA GTGACCAACT GGCCCAAGCC ATCAGTGACC ACTATCCAGT GGAGGTGATG
     1561 CTGAAGTGA
//
```

Fig. 15(C)

```
                9          18          27          36          45          54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63          72          81          90          99         108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117         126         135         144         153         162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171         180         189         198         207         216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225         234         243         252         261         270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279         288         297         306         315         324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333         342         351         360         369         378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387         396         405         414         423         432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441         450         459         468         477         486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495         504         513         522         531         540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549         558         567         576         585         594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G
```

Fig. 15(D)
(Sheet 1 of 3)

```
        603         612         621         630         639         648
CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

        657         666         675         684         693         702
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC TGT GTG GAG TGC CCA CCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   C   V   E   C   P   P

        765         774         783         792         801         810
TGC CCA GCA CCT GAA GGC GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 C   P   A   P   E   G   G   L   K   I   A   A   F   N   I   Q   T   F

        819         828         837         846         855         864
GGG GAG ACC AAG ATG TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   E   T   K   M   S   N   A   T   L   V   S   Y   I   V   Q   I   L

        873         882         891         900         909         918
AGC CGC TAC GAC ATC GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   R   Y   D   I   A   L   V   Q   E   V   R   D   S   H   L   T   A

        927         936         945         954         963         972
GTG GGG AAG CTG CTG GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   G   K   L   L   D   N   L   N   Q   D   A   P   D   T   Y   H   Y

        981         990         999        1008        1017        1026
GTG GTC AGT GAG CCA CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   V   S   E   P   L   G   R   N   S   Y   K   E   R   Y   L   F   V

       1035        1044        1053        1062        1071        1080
TAC AGG CCT GAC CAG GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Y   R   P   D   Q   V   S   A   V   D   S   Y   Y   Y   D   D   G   C

       1089        1098        1107        1116        1125        1134
GAG CCC TGC GGG AAC GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   P   C   G   N   D   T   F   N   R   E   P   A   I   V   R   F   F

       1143        1152        1161        1170        1179        1188
TCC CGG TTC ACA GAG GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GCC CCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   R   F   T   E   V   R   E   F   A   I   V   P   L   H   A   A   P

       1197        1206        1215        1224        1233        1242
```


Fig. 15(D)
(Sheet 2 of 3)

```
GGG GAC GCA GTA GCC GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   D   A   V   A   E   I   D   A   L   Y   D   V   Y   L   D   V   Q

       1251        1260        1269        1278        1287        1296
GAG AAA TGG GGC TTG GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   K   W   G   L   E   D   V   M   L   M   G   D   F   N   A   G   C

       1305        1314        1323        1332        1341        1350
AGC TAT GTG AGA CCC TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   Y   V   R   P   S   Q   W   S   S   I   R   L   W   T   S   P   T

       1359        1368        1377        1386        1395        1404
TTC CAG TGG CTG ATC CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 F   Q   W   L   I   P   D   S   A   D   T   T   A   T   P   T   H   C

       1413        1422        1431        1440        1449        1458
GCC TAT GAC AGG ATC GTG GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT GTT CCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   Y   D   R   I   V   V   A   G   M   L   L   R   G   A   V   V   P

       1467        1476        1485        1494        1503        1512
GAC TCG GCT CTT CCC TTT AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC CAA CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   S   A   L   P   F   N   F   Q   A   A   Y   G   L   S   D   Q   L

       1521        1530        1539        1548        1557
GCC CAA GCC ATC AGT GAC CAC TAT CCA GTG GAG GTG ATG CTG AAG TGA 3'
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   Q   A   I   S   D   H   Y   P   V   E   V   M   L   K   *
```

Fig. 15(D)
(Sheet 3 of 3)

70/113

pAS104

```
LOCUS       PAS104.DNA   1560 bp    mRNA            PRI       06-MAR-1995
DEFINITION  Humanised HMFG1 Fab'2 fused to human DNase I (pAS104)
Position 924 G to A by ggg to gag
Linker GR instead of GG (position 777)
ACCESSION
NID
KEYWORDS    DNase I.
SOURCE      DNase I sequence is from assembled oligos (thus modified c/f
MHDNASE1.dna)
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
  AUTHORS   Shak,S., Capon,D.J., Hellmiss,R., Marsters,S.A. and Baker,C.L.
  TITLE     Recombinant human DNase I reduces the viscosity of cystic
fibrosis
            sputum
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 87 (23), 9188-9192 (1990)
  MEDLINE   91067672
BASE COUNT      346 a    468 c    434 g    312 t
ORIGIN

        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCTG TGTGGAGTGC CCACCGTGCC CAGCACCTGA AGGCAGGCTG
      781 AAGATCGCAG CCTTCAACAT CCAGACATTT GGGGAGACCA AGATGTCCAA TGCCACCCTC
      841 GTCAGCTACA TTGTGCAGAT CCTGAGCCGC TACGACATCG CCCTGGTCCA GGAGGTCAGA
      901 GACAGCCACC TGACTGCCGT GGAGAAGCTG CTGGACAACC TCAATCAGGA CGCACCAGAC
      961 ACCTATCACT ACGTGGTCAG TGAGCCACTG GGACGGAACA GCTATAAGGA GCGCTACCTG
     1021 TTCGTGTACA GGCCTGACCA GGTGTCTGCG GTGGACAGCT ACTACTACGA TGATGGCTGC
     1081 GAGCCCTGCG GGAACGACAC CTTCAACCGA GAGCCAGCCA TTGTCAGGTT CTTCTCCCGG
     1141 TTCACAGAGG TCAGGGAGTT TGCCATTGTT CCCCTGCATG CGGCCCCGGG GGACGCAGTA
     1201 GCCGAGATCG ACGCTCTCTA TGACGTCTAC CTGGATGTCC AAGAGAAATG GGGCTTGGAG
     1261 GACGTCATGT TGATGGGCGA CTTCAATGCG GGCTGCAGCT ATGTGAGACC CTCCCAGTGG
     1321 TCATCCATCC GCCTGTGGAC AAGCCCCACC TTCCAGTGGC TGATCCCCGA CAGCGCTGAC
     1381 ACCACAGCTA CACCCACGCA CTGTGCCTAT GACAGGATCG TGGTTGCAGG GATGCTGCTC
     1441 CGAGGGGCCG TTGTTCCCGA CTCGGCTCTT CCCTTTAACT TCCAGGCTGC CTATGGCCTG
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//
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Fig. 16(A)

71/113

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LOCUS       FDDNASE104   1560 BP SS-DNA             SYN       25-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
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     frag            721..792
                     /note="1 to 72 of 104linker"
     frag            join(721..>774,<776..792)
                     /note="1 to 72 of 103linker [Split]"
     frag            join(721..>771,<772..>774,<776..792)
                     /note="1 to 78 of 102linker [Split]"
BASE COUNT      346 A    467 C    434 G    313 T      0 OTHER
ORIGIN      -
        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCTG TGTCGAGTGT CCACCGTGTC CAGCACCAGA GGGCAGGCTG
      781 AAGATCGCAG CCTTCAACAT CCAGACATTT GGGGAGACCA AGATGTCCAA TGCCACCCTC
      841 GTCAGCTACA TTGTGCAGAT CCTGAGCCGC TACGACATCG CCCTGGTCCA GGAGGTCAGA
      901 GACAGCCACC TGACTGCCGT GGAGAAGCTG CTGGACAACC TCAATCAGGA CGCACCAGAC
      961 ACCTATCACT ACGTGGTCAG TGAGCCACTG GGACGGAACA GCTATAAGGA GCGCTACCTG
     1021 TTCGTGTACA GGCCTGACCA GGTGTCTGCG GTGGACAGCT ACTACTACGA TGATGGCTGC
     1081 GAGCCCTGCG GGAACGACAC CTTCAACCGA GAGCCAGCCA TTGTCAGGTT CTTCTCCCGG
     1141 TTCACAGAGG TCAGGGAGTT TGCCATTGTT CCCCTGCATG CGGCCCCGGG GGACGCAGTA
     1201 GCCGAGATCG ACGCTCTCTA TGACGTCTAC CTGGATGTCC AAGAGAAATG GGGCTTGGAG
     1261 GACGTCATGT TGATGGGCGA CTTCAATGCG GGCTGCAGCT ATGTGAGACC CTCCCAGTGG
     1321 TCATCCATCC GCCTGTGGAC AAGCCCCACC TTCCAGTGGC TGATCCCCGA CAGCGCTGAC
     1381 ACCACAGCTA CACCCACGCA CTGTGCCTAT GACAGGATCG TGGTTGCAGG GATGCTGCTC
     1441 CGAGGGGCCG TTGTTCCCGA CTCGGCTCTT CCCTTTAACT TCCAGGCTGC CTATGGCCTG
     1501 AGTGACCAAC TGGCCCAAGC CATCAGTGAC CACTATCCAG TGGAGGTGAT GCTGAAGTGA
//
```

Fig. 16(B)

72/113

```
                9          18          27          36          45          54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63          72          81          90          99         108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117         126         135         144         153         162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171         180         189         198         207         216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225         234         243         252         261         270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279         288         297         306         315         324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333         342         351         360         369         378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387         396         405         414         423         432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441         450         459         468         477         486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495         504         513         522         531         540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549         558         567         576         585         594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G
```

Fig. 16(C)
(Sheet 1 of 3)

```
        603         612         621         630         639         648
CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

        657         666         675         684         693         702
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC TGT GTG GAG TGC CCA CCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   C   V   E   C   P   P

        765         774         783         792         801         810
TGC CCA GCA CCT GAA GGC AGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 C   P   A   P   E   G   R   L   K   I   A   A   F   N   I   Q   T   F

        819         828         837         846         855         864
GGG GAG ACC AAG ATG TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   E   T   K   M   S   N   A   T   L   V   S   Y   I   V   Q   I   L

        873         882         891         900         909         918
AGC CGC TAC GAC ATC GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   R   Y   D   I   A   L   V   Q   E   V   R   D   S   H   L   T   A

        927         936         945         954         963         972
GTG GAG AAG CTG CTG GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   K   L   L   D   N   L   N   Q   D   A   P   D   T   Y   H   Y

        981         990         999        1008        1017        1026
GTG GTC AGT GAG CCA CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   V   S   E   P   L   G   R   N   S   Y   K   E   R   Y   L   F   V

       1035        1044        1053        1062        1071        1080
TAC AGG CCT GAC CAG GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Y   R   P   D   Q   V   S   A   V   D   S   Y   Y   Y   D   D   G   C

       1089        1098        1107        1116        1125        1134
GAG CCC TGC GGG AAC GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   P   C   G   N   D   T   F   N   R   E   P   A   I   V   R   F   F

       1143        1152        1161        1170        1179        1188
TCC CGG TTC ACA GAG GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GGC CCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   R   F   T   E   V   R   E   F   A   I   V   P   L   H   A   A   P

       1197        1206        1215        1224        1233        1242
```

Fig. 16(C)

(Sheet 2 of 3)

```
GGG GAC GCA GTA GCC GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   D   A   V   A   E   I   D   A   L   Y   D   V   Y   L   D   V   Q

       1251        1260        1269        1278        1287        1296
GAG AAA TGG GGC TTG GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   K   W   G   L   E   D   V   M   L   M   G   D   F   N   A   G   C

       1305        1314        1323        1332        1341        1350
AGC TAT GTG AGA CCC TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   Y   V   R   P   S   Q   W   S   S   I   R   L   W   T   S   P   T

       1359        1368        1377        1386        1395        1404
TTC CAG TGG CTG ATC CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 F   Q   W   L   I   P   D   S   A   D   T   T   A   T   P   T   H   C

       1413        1422        1431        1440        1449        1458
GCC TAT GAC AGG ATC GTG GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT GTT CCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   Y   D   R   I   V   V   A   G   M   L   L   R   G   A   V   V   P

       1467        1476        1485        1494        1503        1512
GAC TCG GCT CTT CCC TTT AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC CAA CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   S   A   L   P   F   N   F   Q   A   A   Y   G   L   S   D   Q   L

       1521        1530        1539        1548        1557
GCC CAA GCC ATC AGT GAC CAC TAT CCA GTG GAG GTG ATG CTG AAG TGA 3'
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   Q   A   I   S   D   H   Y   P   V   E   V   M   L   K   *
```

Fig. 16(C)
(Sheet 3 of 3)

75/113

pAS105

```
LOCUS       PAS105.DNA   1578 bp    mRNA            PRI       06-MAR-1995
DEFINITION  Humanised HMFG1 Fab'2 fused to human DNase I with SV40
NLS(pAS105)
ACCESSION
NID
KEYWORDS    DNase I.
SOURCE      DNase I sequence is from assembled oligos (thus modified c/f
MHDNASE1.dna)
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
  AUTHORS   Shak,S., Capon,D.J., Hellmiss,R., Marsters,S.A. and Baker,C.L.
  TITLE     Recombinant human DNase I reduces the viscosity of cystic
fibrosis
            sputum
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 87 (23), 9188-9192 (1990)
  MEDLINE   91067672
BASE COUNT      353 a    473 c    442 g    310 t
ORIGIN

        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCCC ACCGTGCCCA GCACCTGAAG GCGGGCTGAA GATCGCAGCC
      781 TTCAACATCC AGACATTTGG GGAGACCAAG ATGTCCAATG CCACCCTCGT CAGCTACATT
      841 GTGCAGATCC TGAGCCGCTA CGACATCGCC CTGGTCCAGG AGGTCAGAGA CAGCCACCTG
      901 ACTGCCGTGG GGAAGCTGCT GGACAACCTC AATCAGGACG CACCAGACAC CTATCACTAC
      961 GTGGTCAGTG AGCCACTGGG ACGGAACAGC TATAAGGAGC GCTACCTGTT CGTGTACAGG
     1021 CCTGACCAGG TGTCTGCGGT GGACAGCTAC TACTACGATG ATGGCTGCGA GCCCTGCGGG
     1081 AACGACACCT TCAACCGAGA GCCAGCCATT GTCAGGTTCT TCTCCCGGTT CACAGAGGTC
     1141 AGGGAGTTTG CCATTGTTCC CCTGCATGCG GCCCCGGGGG ACGCAGTAGC CGAGATCGAC
     1201 GCTCTCTATG ACGTCTACCT GGATGTCCAA GAGAAATGGG GCTTGGAGGA CGTCATGTTG
     1261 ATGGGCGACT TCAATGCGGG CTGCAGCTAT GTGAGACCCT CCCAGTGGTC ATCCATCCGC
     1321 CTGTGGACAA GCCCCACCTT CCAGTGGCTG ATCCCCGACA GCGCTGACAC CACAGCTACA
     1381 CCCACGCACT GTGCCTATGA CAGGATCGTG GTTGCAGGGA TGCTGCTCCG AGGGGCCGTT
     1441 GTTCCCGACT CGGCTCTTCC CTTTAACTTC CAGGCTGCCT ATGGCCTGAG TGACCAACTG
     1501 GCCCAAGCCA TCAGTGACCA CTATCCAGTG GAGGTGATGC TGAAGGGGGG CGGACCCAAA
     1561 AAGAAGCGCA AGGTTTGA
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NLS

Fig. 17(A)

76/113

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LOCUS       FDDNASE105   1578 BP SS-DNA             SYN       25-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
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     frag            721..780
                     /note="1 to 60 of 101/105linker"
     frag            join(721..>735,<736..>759,<760..>780)
                     /note="1 to 80 of 102linker [Split]"
BASE COUNT      353 A    471 C    443 G    311 T      0 OTHER
ORIGIN      -
        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGTCC ACCGTGTCCA GCACCAGAGG GCGGGCTGAA GATCGCAGCC
      781 TTCAACATCC AGACATTTGG GGAGACCAAG ATGTCCAATG CCACCCTCGT CAGCTACATT
      841 GTGCAGATCC TGAGCCGCTA CGACATCGCC CTGGTCCAGG AGGTCAGAGA CAGCCACCTG
      901 ACTGCCGTGG GGAAGCTGCT GGACAACCTC AATCAGGACG CACCAGACAC CTATCACTAC
      961 GTGGTCAGTG AGCCACTGGG ACGGAACAGC TATAAGGAGC GCTACCTGTT CGTGTACAGG
     1021 CCTGACCAGG TGTCTGCGGT GGACAGCTAC TACTACGATG ATGGCTGCGA GCCCTGCGGG
     1081 AACGACACCT TCAACCGAGA GCCAGCCATT GTCAGGTTCT TCTCCCGGTT CACAGAGGTC
     1141 AGGGAGTTTG CCATTGTTCC CCTGCATGCG GCCCCGGGGG ACGCAGTAGC CGAGATCGAC
     1201 GCTCTCTATG ACGTCTACCT GGATGTCCAA GAGAAATGGG GCTTGGAGGA CGTCATGTTG
     1261 ATGGGCGACT TCAATGCGGG CTGCAGCTAT GTGAGACCCT CCCAGTGGTC ATCCATCCGC
     1321 CTGTGGACAA GCCCCACCTT CCAGTGGCTG ATCCCCGACA GCGCTGACAC CACAGCTACA
     1381 CCCACGCACT GTGCCTATGA CAGGATCGTG GTTGCAGGGA TGCTGCTCCG AGGGGCCGTT
     1441 GTTCCCGACT CGGCTCTTCC CTTTAACTTC CAGGCTGCCT ATGGCCTGAG TGACCAACTG
     1501 GCCCAAGCCA TCAGTGACCA CTATCCAGTG GAGGTGATGC TGAAGGGGGG CGGACCCAAA
     1561 AAGAAGCGCA AGGTTTGA
//
```

Fig. 17(B)

77/113

```
LOCUS       FDDNASE105   1587 BP SS-DNA             SYN       29-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
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                     /note="1 to 1578 of PAS105.dna [Split]"
     frag            730..789
                     /note="1 to 60 of 101/105linker"
     frag            join(730..>744,<745..>768,<769..>789)
                     /note="1 to 80 of 102linker [Split]"
BASE COUNT      354 A    477 C    445 G    311 T      0 OTHER
ORIGIN      -
        1 GCCGCCACCA TGGGATGGAG CTGTATCATC CTCTTCTTGG TAGCAACAGC TACAGGTGTC
       61 CACTCCCAGG TGCAGCTGGT GCAGTCTGGG GCAGAGGTGA AAAAGCCTGG GGCCTCAGTG
      121 AAGGTGTCCT GCAAGGCTTC TGGCTACACC TTCAGTGCCT ACTGGATAGA GTGGGTGCGC
      181 CAGGCTCCAG GAAAGGGCCT CGAGTGGGTC GGAGAGATTT TACCTGGAAG TAATAATTCT
      241 AGATACAATG AGAAGTTCAA GGGCCGAGTG ACAGTCACTA GAGACACATC CACAAACACA
      301 GCCTACATGG AGCTCAGCAG CCTGAGGTCT GAGGACACAG CCGTCTATTA CTGTGCAAGA
      361 TCCTACGACT TTGCCTGGTT TGCTTACTGG GGCCAAGGGA CTCTGGTCAC AGTCTCCTCA
      421 GCCTCCACCA AGGGCCCATC GGTCTTCCCC CTGGCACCCT CCTCCAAGAG CACCTCTGGG
      481 GGCACAGCGG CCCTGGGCTG CCTGGTCAAG GACTACTTCC CCGAACCGGT GACGGTGTCG
      541 TGGAACTCAG GCGCCCTGAC CAGCGGCGTG CACACCTTCC CGGCTGTCCT ACAGTCCTCA
      601 GGACTCTACT CCCTCAGCAG CGTGGTGACC GTGCCCTCCA GCAGCTTGGG CACCCAGACC
      661 TACATCTGCA ACGTGAATCA CAAGCCCAGC AACACCAAGG TGGACAAGAA AGTTGAGCCC
      721 AAATCTTGTG ACAAAACTCA CACATGTCCA CCGTGTCCAG CACCAGAGGG CGGGCTGAAG
      781 ATCGCAGCCT TCAACATCCA GACATTTGGG GAGACCAAGA TGTCCAATGC CACCCTCGTC
      841 AGCTACATTG TGCAGATCCT GAGCCGCTAC GACATCGCCC TGGTCCAGGA GGTCAGAGAC
      901 AGCCACCTGA CTGCCGTGGG GAAGCTGCTG GACAACCTCA ATCAGGACGC ACCAGACACC
      961 TATCACTACG TGGTCAGTGA GCCACTGGGA CGGAACAGCT ATAAGGAGCG CTACCTGTTC
     1021 GTGTACAGGC CTGACCAGGT GTCTGCGGTG GACAGCTACT ACTACGATGA TGGCTGCGAG
     1081 CCCTGCGGGA ACGACACCTT CAACCGAGAG CCAGCCATTG TCAGGTTCTT CTCCCGGTTC
     1141 ACAGAGGTCA GGGAGTTTGC CATTGTTCCC CTGCATGCGG CCCCGGGGGA CGCAGTAGCC
     1201 GAGATCGACG CTCTCTATGA CGTCTACCTG GATGTCCAAG AGAAATGGGG CTTGGAGGAC
     1261 GTCATGTTGA TGGGCGACTT CAATGCGGGC TGCAGCTATG TGAGACCCTC CCAGTGGTCA
     1321 TCCATCCGCC TGTGGACAAG CCCCACCTTC CAGTGGCTGA TCCCCGACAG CGCTGACACC
     1381 ACAGCTACAC CCACGCACTG TGCCTATGAC AGGATCGTGG TTGCAGGGAT GCTGCTCCGA
     1441 GGGGCCGTTG TTCCCGACTC GGCTCTTCCC TTTAACTTCC AGGCTGCCTA TGGCCTGAGT
     1501 GACCAACTGG CCCAAGCCAT CAGTGACCAC TATCCAGTGG AGGTGATGCT GAAGGGGGGC
     1561 GGACCCAAAA AGAAGCGCAA GGTTTGA
//
```

Fig. 17(C)

```
                9          18          27          36          45          54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63          72          81          90          99         108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117         126         135         144         153         162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171         180         189         198         207         216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225         234         243         252         261         270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279         288         297         306         315         324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333         342         351         360         369         378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387         396         405         414         423         432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441         450         459         468         477         486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495         504         513         522         531         540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549         558         567         576         585         594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G
```


Fig. 17(D)
(Sheet 1 of 3)

```
        603         612         621         630         639         648
CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

        657         666         675         684         693         702
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC CCA CCG TGC CCA GCA CCT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   P   P   C   P   A   P

        765         774         783         792         801         810
GAA GGC GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT GGG GAG ACC AAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   G   G   L   K   I   A   A   F   N   I   Q   T   F   G   E   T   K

        819         828         837         846         855         864
ATG TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG AGC CGC TAC GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 M   S   N   A   T   L   V   S   Y   I   V   Q   I   L   S   R   Y   D

        873         882         891         900         909         918
ATC GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC GTG GGG AAG CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   A   L   V   Q   E   V   R   D   S   H   L   T   A   V   G   K   L

        927         936         945         954         963         972
CTG GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC GTG GTC AGT GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   D   N   L   N   Q   D   A   P   D   T   Y   H   Y   V   V   S   E

        981         990         999        1008        1017        1026
CCA CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG TAC AGG CCT GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   L   G   R   N   S   Y   K   E   R   Y   L   F   V   Y   R   P   D

       1035        1044        1053        1062        1071        1080
CAG GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC GAG CCC TGC GGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Q   V   S   A   V   D   S   Y   Y   Y   D   D   G   C   E   P   C   G

       1089        1098        1107        1116        1125        1134
AAC GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC TCC CGG TTC ACA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 N   D   T   F   N   R   E   P   A   I   V   R   F   F   S   R   F   T

       1143        1152        1161        1170        1179        1188
GAG GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GCC CCG GGG GAC GCA GTA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   V   R   E   F   A   I   V   P   L   H   A   A   P   G   D   A   V

       1197        1206        1215        1224        1233        1242
```

Fig. 17(D)

(Sheet 2 of 3)

80/113

```
GCC GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA GAG AAA TGG GGC
 A   E   I   D   A   L   Y   D   V   Y   L   D   V   Q   E   K   W   G

      1251        1260        1269        1278        1287        1296
TTG GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC AGC TAT GTG AGA
 L   E   D   V   M   L   M   G   D   F   N   A   G   C   S   Y   V   R

      1305        1314        1323        1332        1341        1350
CCC TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC TTC CAG TGG CTG
 P   S   Q   W   S   S   I   R   L   W   T   S   P   T   F   Q   W   L

      1359        1368        1377        1386        1395        1404
ATC CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT GCC TAT GAC AGG
 I   P   D   S   A   D   T   T   A   T   P   T   H   C   A   Y   D   R

      1413        1422        1431        1440        1449        1458
ATC GTG GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT GTT CCC GAC TCG GCT CTT
 I   V   V   A   G   M   L   L   R   G   A   V   V   P   D   S   A   L

      1467        1476        1485        1494        1503        1512
CCC TTT AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC CAA CTG GCC CAA GCC ATC
 P   F   N   F   Q   A   A   Y   G   L   S   D   Q   L   A   Q   A   I

      1521        1530        1539        1548        1557        1566
AGT GAC CAC TAT CCA GTG GAG GTG ATG CTG AAG GGG GGC GGA CCC AAA AAG AAG
 S   D   H   Y   P   V   E   V   M   L   K   G   G   G   P   K   K   K

      1575
CGC AAG GTT TGA 3'
 R   K   V   *
```

Fig. 17(D)
(Sheet 3 of 3)

81/113

pAS106

```
LOCUS       PAS106.DNA   1596 bp    mRNA            PRI       06-MAR-1995
DEFINITION  Humanised HMFG1 Fab'2 fused to human DNase I with SV40
NLS(pAS106)
ACCESSION
NID
KEYWORDS    DNase I.
SOURCE      DNase I sequence is from assembled oligos (thus modified c/f
MHDNASE1.dna)
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
  AUTHORS   Shak,S., Capon,D.J., Hellmiss,R., Marsters,S.A. and Baker,C.L.
  TITLE     Recombinant human DNase I reduces the viscosity of cystic
fibrosis
            sputum
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 87 (23), 9188-9192 (1990)
  MEDLINE   91067672
BASE COUNT      355 a    475 c    452 g    314 t
ORIGIN

        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCTG TGTGGAGTGC CCACCGTGCC CAGCACCTGA AGGGAGCGGC
      781 GGGCTGAAGA TCGCAGCCTT CAACATCCAG ACATTTGGGG AGACCAAGAT GTCCAATGCC
      841 ACCCTCGTCA GCTACATTGT GCAGATCCTG AGCCGCTACG ACATCGCCCT GGTCCAGGAG
      901 GTCAGAGACA GCCACCTGAC TGCCGTGGGG AAGCTGCTGG ACAACCTCAA TCAGGACGCA
      961 CCAGACACCT ATCACTACGT GGTCAGTGAG CCACTGGGAC GGAACAGCTA TAAGGAGCGC
     1021 TACCTGTTCG TGTACAGGCC TGACCAGGTG TCTGCGGTGG ACAGCTACTA CTACGATGAT
     1081 GGCTGCGAGC CCTGCGGGAA CGACACCTTC AACCGAGAGC CAGCCATTGT CAGGTTCTTC
     1141 TCCCGGTTCA CAGAGGTCAG GGAGTTTGCC ATTGTTCCCC TGCATGCGGC CCCGGGGGAC
     1201 GCAGTAGCCG AGATCGACGC TCTCTATGAC GTCTACCTGG ATGTCCAAGA GAAATGGGGC
     1261 TTGGAGGACG TCATGTTGAT GGGCGACTTC AATGCGGGCT GCAGCTATGT GAGACCCTCC
     1321 CAGTGGTCAT CCATCCGCCT GTGGACAAGC CCCACCTTCC AGTGGCTGAT CCCCGACAGC
     1381 GCTGACACCA CAGCTACACC CACGCACTGT GCCTATGACA GGATCGTGGT TGCAGGGATG
     1441 CTGCTCCGAG GGGCCGTTGT TCCCGACTCG GCTCTTCCCT TTAACTTCCA GGCTGCCTAT
     1501 GGCCTGAGTG ACCAACTGGC CCAAGCCATC AGTGACCACT ATCCAGTGGA GGTGATGCTG
     1561 AAGGGGGGCG GACCCAAAAA GAAGCGCAAG GTTTGA
//
```

NLS

Fig. 18(A)

82/113

```
LOCUS       FDDNASE106   1596 BP SS-DNA             SYN       25-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
     frag            join(1..>720,<799..1596)
                     /note="1 to 1596 of PAS106.dna [Split]"
     frag            721..798
                     /note="1 to 78 of 102/106linker"
BASE COUNT      355 A    474 C    452 G    315 T      0 OTHER
ORIGIN      -
        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCTG TGTCGAGTGT CCACCGTGTC CAGCACCAGA GGGGAGCGGC
      781 GGGCTGAAGA TCGCAGCCTT CAACATCCAG ACATTTGGGG AGACCAAGAT GTCCAATGCC
      841 ACCCTCGTCA GCTACATTGT GCAGATCCTG AGCCGCTACG ACATCGCCCT GGTCCAGGAG
      901 GTCAGAGACA GCCACCTGAC TGCCGTGGGG AAGCTGCTGG ACAACCTCAA TCAGGACGCA
      961 CCAGACACCT ATCACTACGT GGTCAGTGAG CCACTGGGAC GGAACAGCTA TAAGGAGCGC
     1021 TACCTGTTCG TGTACAGGCC TGACCAGGTG TCTGCGGTGG ACAGCTACTA CTACGATGAT
     1081 GGCTGCGAGC CCTGCGGGAA CGACACCTTC AACCGAGAGC CAGCCATTGT CAGGTTCTTC
     1141 TCCCGGTTCA CAGAGGTCAG GGAGTTTGCC ATTGTTCCCC TGCATGCGGC CCCGGGGGAC
     1201 GCAGTAGCCG AGATCGACGC TCTCTATGAC GTCTACCTGG ATGTCCAAGA GAAATGGGGC
     1261 TTGGAGGACG TCATGTTGAT GGGCGACTTC AATGCGGGCT GCAGCTATGT GAGACCCTCC
     1321 CAGTGGTCAT CCATCCGCCT GTGGACAAGC CCCACCTTCC AGTGGCTGAT CCCCGACAGC
     1381 GCTGACACCA CAGCTACACC CACGCACTGT GCCTATGACA GGATCGTGGT TGCAGGGATG
     1441 CTGCTCCGAG GGGCCGTTGT TCCCGACTCG GCTCTTCCCT TTAACTTCCA GGCTGCCTAT
     1501 GGCCTGAGTG ACCAACTGGC CCAAGCCATC AGTGACCACT ATCCAGTGGA GGTGATGCTG
     1561 AAGGGGGGCG GACCCAAAAA GAAGCGCAAG GTTTGA
//
```

Fig. 18(B)

83/113

```
LOCUS       FDDNASE106   1605 BP SS-DNA             SYN       29-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
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                     /note="1 to 1596 of FdDNase106correct"
     frag            join(10..>729,<808..1605)
                     /note="1 to 1596 of PAS106.dna [Split]"
     frag            730..807
                     /note="1 to 78 of 102/106linker"
BASE COUNT      356 A    480 C    454 G    315 T      0 OTHER
ORIGIN      -
        1 GCCGCCACCA TGGGATGGAG CTGTATCATC CTCTTCTTGG TAGCAACAGC TACAGGTGTC
       61 CACTCCCAGG TGCAGCTGGT GCAGTCTGGG GCAGAGGTGA AAAAGCCTGG GGCCTCAGTG
      121 AAGGTGTCCT GCAAGGCTTC TGGCTACACC TTCAGTGCCT ACTGGATAGA GTGGGTGCGC
      181 CAGGCTCCAG GAAAGGGCCT CGAGTGGGTC GGAGAGATTT TACCTGGAAG TAATAATTCT
      241 AGATACAATG AGAAGTTCAA GGGCCGAGTG ACAGTCACTA GAGACACATC CACAAACACA
      301 GCCTACATGG AGCTCAGCAG CCTGAGGTCT GAGGACACAG CCGTCTATTA CTGTGCAAGA
      361 TCCTACGACT TTGCCTGGTT TGCTTACTGG GGCCAAGGGA CTCTGGTCAC AGTCTCCTCA
      421 GCCTCCACCA AGGGCCCATC GGTCTTCCCC CTGGCACCCT CCTCCAAGAG CACCTCTGGG
      481 GGCACAGCGG CCCTGGGCTG CCTGGTCAAG GACTACTTCC CCGAACCGGT GACGGTGTCG
      541 TGGAACTCAG GCGCCCTGAC CAGCGGCGTG CACACCTTCC CGGCTGTCCT ACAGTCCTCA
      601 GGACTCTACT CCCTCAGCAG CGTGGTGACC GTGCCCTCCA GCAGCTTGGG CACCCAGACC
      661 TACATCTGCA ACGTGAATCA CAAGCCCAGC AACACCAAGG TGGACAAGAA AGTTGAGCCC
      721 AAATCTTGTG ACAAAACTCA CACATGCTGT GTCGAGTGTC CACCGTGTCC AGCACCAGAG
      781 GGGAGCGGCG GGCTGAAGAT CGCAGCCTTC AACATCCAGA CATTTGGGGA GACCAAGATG
      841 TCCAATGCCA CCCTCGTCAG CTACATTGTG CAGATCCTGA GCCGCTACGA CATCGCCCTG
      901 GTCCAGGAGG TCAGAGACAG CCACCTGACT GCCGTGGGGA AGCTGCTGGA CAACCTCAAT
      961 CAGGACGCAC CAGACACCTA TCACTACGTG GTCAGTGAGC CACTGGGACG GAACAGCTAT
     1021 AAGGAGCGCT ACCTGTTCGT GTACAGGCCT GACCAGGTGT CTGCGGTGGA CAGCTACTAC
     1081 TACGATGATG GCTGCGAGCC CTGCGGGAAC GACACCTTCA ACCGAGAGCC AGCCATTGTC
     1141 AGGTTCTTCT CCCGGTTCAC AGAGGTCAGG GAGTTTGCCA TTGTTCCCCT GCATGCGGCC
     1201 CCGGGGGACG CAGTAGCCGA GATCGACGCT CTCTATGACG TCTACCTGGA TGTCCAAGAG
     1261 AAATGGGGCT TGGAGGACGT CATGTTGATG GGCGACTTCA ATGCGGGCTG CAGCTATGTG
     1321 AGACCCTCCC AGTGGTCATC CATCCGCCTG TGGACAAGCC CCACCTTCCA GTGGCTGATC
     1381 CCCGACAGCG CTGACACCAC AGCTACACCC ACGCACTGTG CCTATGACAG GATCGTGGTT
     1441 GCAGGGATGC TGCTCCGAGG GGCCGTTGTT CCCGACTCGG CTCTTCCCTT TAACTTCCAG
     1501 GCTGCCTATG GCCTGAGTGA CCAACTGGCC CAAGCCATCA GTGACCACTA TCCAGTGGAG
     1561 GTGATGCTGA AGGGGGGCGG ACCCAAAAAG AAGCGCAAGG TTTGA
//
```

Fig. 18(C)

```
          9          18          27          36          45          54
ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

         63          72          81          90          99         108
TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

        117         126         135         144         153         162
GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

        171         180         189         198         207         216
TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

        225         234         243         252         261         270
GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

        279         288         297         306         315         324
AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

        333         342         351         360         369         378
GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

        387         396         405         414         423         432
TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

        441         450         459         468         477         486
GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

        495         504         513         522         531         540
GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

        549         558         567         576         585         594
GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G
```

Fig. 18(C)

(Sheet 1 of 3)

85/113

```
        603         612         621         630         639         648
CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

        657         666         675         684         693         702
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC TGT GTG GAG TGC CCA CCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   C   V   E   C   P   P

        765         774         783         792         801         810
TGC CCA GCA CCT GAA GGG AGC GGC GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 C   P   A   P   E   G   S   G   G   L   K   I   A   A   F   N   I   Q

        819         828         837         846         855         864
ACA TTT GGG GAG ACC AAG ATG TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   F   G   E   T   K   M   S   N   A   T   L   V   S   Y   I   V   Q

        873         882         891         900         909         918
ATC CTG AGC CGC TAC GAC ATC GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 I   L   S   R   Y   D   I   A   L   V   Q   E   V   R   D   S   H   L

        927         936         945         954         963         972
ACT GCC GTG GGG AAG CTG CTG GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   A   V   G   K   L   L   D   N   L   N   Q   D   A   P   D   T   Y

        981         990         999        1008        1017        1026
CAC TAC GTG GTC AGT GAG CCA CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 H   Y   V   V   S   E   P   L   G   R   N   S   Y   K   E   R   Y   L

       1035        1044        1053        1062        1071        1080
TTC GTG TAC AGG CCT GAC CAG GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 F   V   Y   R   P   D   Q   V   S   A   V   D   S   Y   Y   Y   D   D

       1089        1098        1107        1116        1125        1134
GGC TGC GAG CCC TGC GGG AAC GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   C   E   P   C   G   N   D   T   F   N   R   E   P   A   I   V   R

       1143        1152        1161        1170        1179        1188
TTC TTC TCC CGG TTC ACA GAG GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 F   F   S   R   F   T   E   V   R   E   F   A   I   V   P   L   H   A

       1197        1206        1215        1224        1233        1242
```

Fig. 18(C)

(Sheet 2 of 3)

86/113

```
GCC CCG GGG GAC GCA GTA GCC GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 A   P   G   D   A   V   A   E   I   D   A   L   Y   D   V   Y   L   D

       1251        1260        1269        1278        1287        1296
GTC CAA GAG AAA TGG GGC TTG GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   Q   E   K   W   G   L   E   D   V   M   L   M   G   D   F   N   A

       1305        1314        1323        1332        1341        1350
GGC TGC AGC TAT GTG AGA CCC TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   C   S   Y   V   R   P   S   Q   W   S   S   I   R   L   W   T   S

       1359        1368        1377        1386        1395        1404
CCC ACC TTC CAG TGG CTG ATC CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 P   T   F   Q   W   L   I   P   D   S   A   D   T   T   A   T   P   T

       1413        1422        1431        1440        1449        1458
CAC TGT GCC TAT GAC AGG ATC GTG GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 H   C   A   Y   D   R   I   V   V   A   G   M   L   L   R   G   A   V

       1467        1476        1485        1494        1503        1512
GTT CCC GAC TCG GCT CTT CCC TTT AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   P   D   S   A   L   P   F   N   F   Q   A   A   Y   G   L   S   D

       1521        1530        1539        1548        1557        1566
CAA CTG GCC CAA GCC ATC AGT GAC CAC TAT CCA GTG GAG GTG ATG CTG AAG GGG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Q   L   A   Q   A   I   S   D   H   Y   P   V   E   V   M   L   K   G

       1575        1584        1593
GGC GGA CCC AAA AAG AAG CGC AAG GTT TGA 3'
--- --- --- --- --- --- --- --- --- ---
 G   G   P   K   K   K   R   K   V   *
```

Fig. 18(C)
(Sheet 3 of 3)

87/113

pAS107

```
LOCUS       PAS107.DNA   1590 bp    mRNA            PRI       06-MAR-1995
DEFINITION  Humanised HMFG1 Fab'2 fused to human DNase I with SV40
NLS(pAS107)
ACCESSION
NID
KEYWORDS    DNase I.
SOURCE      DNase I sequence is from assembled oligos (thus modified c/f
MHDNASE1.dna)
  ORGANISM  Homo sapiens
            Eukaryotae; mitochondrial eukaryotes; Metazoa; Chordata;
            Vertebrata; Eutheria; Primates; Catarrhini; Hominidae; Homo.
  AUTHORS   Shak,S., Capon,D.J., Hellmiss,R., Marsters,S.A. and Baker,C.L.
  TITLE     Recombinant human DNase I reduces the viscosity of cystic
fibrosis
            sputum
  JOURNAL   Proc. Natl. Acad. Sci. U.S.A. 87 (23), 9188-9192 (1990)
  MEDLINE   91067672
BASE COUNT      354 a    474 c    448 g    314 t
ORIGIN

        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCTG TGTGGAGTGC CCACCGTGCC CAGCACCTGA AGGCGGGCTG
      781 AAGATCGCAG CCTTCAACAT CCAGACATTT GGGGAGACCA AGATGTCCAA TGCCACCCTC
      841 GTCAGCTACA TTGTGCAGAT CCTGAGCCGC TACGACATCG CCCTGGTCCA GGAGGTCAGA
      901 GACAGCCACC TGACTGCCGT GGGGAAGCTG CTGGACAACC TCAATCAGGA CGCACCAGAC
      961 ACCTATCACT ACGTGGTCAG TGAGCCACTG GGACGGAACA GCTATAAGGA GCGCTACCTG
     1021 TTCGTGTACA GGCCTGACCA GGTGTCTGCG GTGGACAGCT ACTACTACGA TGATGGCTGC
     1081 GAGCCCTGCG GGAACGACAC CTTCAACCGA GAGCCAGCCA TTGTCAGGTT CTTCTCCCGG
     1141 TTCACAGAGG TCAGGGAGTT TGCCATTGTT CCCCTGCATG CGGCCCCGGG GGACGCAGTA
     1201 GCCGAGATCG ACGCTCTCTA TGACGTCTAC CTGGATGTCC AAGAGAAATG GGGCTTGGAG
     1261 GACGTCATGT TGATGGGCGA CTTCAATGCG GGCTGCAGCT ATGTGAGACC CTCCCAGTGG
     1321 TCATCCATCC GCCTGTGGAC AAGCCCCACC TTCCAGTGGC TGATCCCCGA CAGCGCTGAC
     1381 ACCACAGCTA CACCCACGCA CTGTGCCTAT GACAGGATCG TGGTTGCAGG GATGCTGCTC
     1441 CGAGGGGCCG TTGTTCCCGA CTCGGCTCTT CCCTTTAACT TCCAGGCTGC CTATGGCCTG
     1501 AGTGACCAAC TGGCCCAAGC CATCAGTGAC CACTATCCAG TGGAGGTGAT GCTGAAGGGG
     1561 GGCGGACCCA AAAAGAAGCG CAAGGTTTGA
//
```

NLS

Fig. 19(A)

88/113

```
LOCUS       FDDNASE107   1590 BP SS-DNA             SYN       25-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
     frag            join(1..>720,<793..1590)
                     /note="1 to 1590 of PAS107.dna [Split]"
     frag            721..792
                     /note="1 to 72 of 103/107linker"
     frag            join(721..>771,<772..792)
                     /note="1 to 78 of 102linker [Split]"
BASE COUNT       354 A    473 C    448 G    315 T      0 OTHER
ORIGIN      -
        1 ATGGGATGGA GCTGTATCAT CCTCTTCTTG GTAGCAACAG CTACAGGTGT CCACTCCCAG
       61 GTGCAGCTGG TGCAGTCTGG GGCAGAGGTG AAAAAGCCTG GGGCCTCAGT GAAGGTGTCC
      121 TGCAAGGCTT CTGGCTACAC CTTCAGTGCC TACTGGATAG AGTGGGTGCG CCAGGCTCCA
      181 GGAAAGGGCC TCGAGTGGGT CGGAGAGATT TTACCTGGAA GTAATAATTC TAGATACAAT
      241 GAGAAGTTCA AGGGCCGAGT GACAGTCACT AGAGACACAT CCACAAACAC AGCCTACATG
      301 GAGCTCAGCA GCCTGAGGTC TGAGGACACA GCCGTCTATT ACTGTGCAAG ATCCTACGAC
      361 TTTGCCTGGT TTGCTTACTG GGGCCAAGGG ACTCTGGTCA CAGTCTCCTC AGCCTCCACC
      421 AAGGGCCCAT CGGTCTTCCC CCTGGCACCC TCCTCCAAGA GCACCTCTGG GGGCACAGCG
      481 GCCCTGGGCT GCCTGGTCAA GGACTACTTC CCCGAACCGG TGACGGTGTC GTGGAACTCA
      541 GGCGCCCTGA CCAGCGGCGT GCACACCTTC CCGGCTGTCC TACAGTCCTC AGGACTCTAC
      601 TCCCTCAGCA GCGTGGTGAC CGTGCCCTCC AGCAGCTTGG GCACCCAGAC CTACATCTGC
      661 AACGTGAATC ACAAGCCCAG CAACACCAAG GTGGACAAGA AAGTTGAGCC CAAATCTTGT
      721 GACAAAACTC ACACATGCTG TGTCGAGTGT CCACCGTGTC CAGCACCAGA GGGCGGGCTG
      781 AAGATCGCAG CCTTCAACAT CCAGACATTT GGGGAGACCA AGATGTCCAA TGCCACCCTC
      841 GTCAGCTACA TTGTGCAGAT CCTGAGCCGC TACGACATCG CCCTGGTCCA GGAGGTCAGA
      901 GACAGCCACC TGACTGCCGT GGGGAAGCTG CTGGACAACC TCAATCAGGA CGCACCAGAC
      961 ACCTATCACT ACGTGGTCAG TGAGCCACTG GGACGGAACA GCTATAAGGA GCGCTACCTG
     1021 TTCGTGTACA GGCCTGACCA GGTGTCTGCG GTGGACAGCT ACTACTACGA TGATGGCTGC
     1081 GAGCCCTGCG GGAACGACAC CTTCAACCGA GAGCCAGCCA TTGTCAGGTT CTTCTCCCGG
     1141 TTCACAGAGG TCAGGGAGTT TGCCATTGTT CCCCTGCATG CGGCCCCGGG GGACGCAGTA
     1201 GCCGAGATCG ACGCTCTCTA TGACGTCTAC CTGGATGTCC AAGAGAAATG GGGCTTGGAG
     1261 GACGTCATGT TGATGGGCGA CTTCAATGCG GGCTGCAGCT ATGTGAGACC CTCCCAGTGG
     1321 TCATCCATCC GCCTGTGGAC AAGCCCCACC TTCCAGTGGC TGATCCCCGA CAGCGCTGAC
     1381 ACCACAGCTA CACCCACGCA CTGTGCCTAT GACAGGATCG TGGTTGCAGG GATGCTGCTC
     1441 CGAGGGGCCG TTGTTCCCGA CTCGGCTCTT CCCTTTAACT TCCAGGCTGC CTATGGCCTG
     1501 AGTGACCAAC TGGCCCAAGC CATCAGTGAC CACTATCCAG TGGAGGTGAT GCTGAAGGGG
     1561 GGCGGACCCA AAAAGAAGCG CAAGGTTTGA
//
```

Fig. 19(B)

89/113

```
LOCUS       FDDNASE107   1599 BP SS-DNA             SYN       29-AUG-2000
DEFINITION  -
ACCESSION   -
KEYWORDS    -
SOURCE      -
FEATURES             Location/Qualifiers
     frag            10..1599
                     /note="1 to 1590 of FdDNase107correct"
     frag            join(10..>729,<802..1599)
                     /note="1 to 1590 of PAS107.dna [Split]"
     frag            730..801
                     /note="1 to 72 of 103/107linker"
     frag            join(730..>780,<781..801)
                     /note="1 to 78 of 102linker [Split]"
BASE COUNT      355 A    479 C    450 G    315 T      0 OTHER
ORIGIN      -
        1 GCCGCCACCA TGGGATGGAG CTGTATCATC CTCTTCTTGG TAGCAACAGC TACAGGTGTC
       61 CACTCCCAGG TGCAGCTGGT GCAGTCTGGG GCAGAGGTGA AAAAGCCTGG GGCCTCAGTG
      121 AAGGTGTCCT GCAAGGCTTC TGGCTACACC TTCAGTGCCT ACTGGATAGA GTGGGTGCGC
      181 CAGGCTCCAG GAAAGGGCCT CGAGTGGGTC GGAGAGATTT TACCTGGAAG TAATAATTCT
      241 AGATACAATG AGAAGTTCAA GGGCCGAGTG ACAGTCACTA GAGACACATC CACAAACACA
      301 GCCTACATGG AGCTCAGCAG CCTGAGGTCT GAGGACACAG CCGTCTATTA CTGTGCAAGA
      361 TCCTACGACT TTGCCTGGTT TGCTTACTGG GGCCAAGGGA CTCTGGTCAC AGTCTCCTCA
      421 GCCTCCACCA AGGGCCCATC GGTCTTCCCC CTGGCACCCT CCTCCAAGAG CACCTCTGGG
      481 GGCACAGCGG CCCTGGGCTG CCTGGTCAAG GACTACTTCC CCGAACCGGT GACGGTGTCG
      541 TGGAACTCAG GCGCCCTGAC CAGCGGCGTG CACACCTTCC CGGCTGTCCT ACAGTCCTCA
      601 GGACTCTACT CCCTCAGCAG CGTGGTGACC GTGCCCTCCA GCAGCTTGGG CACCCAGACC
      661 TACATCTGCA ACGTGAATCA CAAGCCCAGC AACACCAAGG TGGACAAGAA AGTTGAGCCC
      721 AAATCTTGTG ACAAAACTCA CACATGCTGT GTCGAGTGTC CACCGTGTCC AGCACCAGAG
      781 GGCGGGCTGA AGATCGCAGC CTTCAACATC CAGACATTTG GGGAGACCAA GATGTCCAAT
      841 GCCACCCTCG TCAGCTACAT TGTGCAGATC CTGAGCCGCT ACGACATCGC CCTGGTCCAG
      901 GAGGTCAGAG ACAGCCACCT GACTGCCGTG GGGAAGCTGC TGGACAACCT CAATCAGGAC
      961 GCACCAGACA CCTATCACTA CGTGGTCAGT GAGCCACTGG GACGGAACAG CTATAAGGAG
     1021 CGCTACCTGT TCGTGTACAG GCCTGACCAG GTGTCTGCGG TGGACAGCTA CTACTACGAT
     1081 GATGGCTGCG AGCCCTGCGG GAACGACACC TTCAACCGAG AGCCAGCCAT TGTCAGGTTC
     1141 TTCTCCCGGT TCACAGAGGT CAGGGAGTTT GCCATTGTTC CCCTGCATGC GGCCCCGGGG
     1201 GACGCAGTAG CCGAGATCGA CGCTCTCTAT GACGTCTACC TGGATGTCCA AGAGAAATGG
     1261 GGCTTGGAGG ACGTCATGTT GATGGGCGAC TTCAATGCGG GCTGCAGCTA TGTGAGACCC
     1321 TCCCAGTGGT CATCCATCCG CCTGTGGACA AGCCCCACCT TCCAGTGGCT GATCCCCGAC
     1381 AGCGCTGACA CCACAGCTAC ACCCACGCAC TGTGCCTATG ACAGGATCGT GGTTGCAGGG
     1441 ATGCTGCTCC GAGGGGCCGT TGTTCCCGAC TCGGCTCTTC CCTTTAACTT CCAGGCTGCC
     1501 TATGGCCTGA GTGACCAACT GGCCCAAGCC ATCAGTGACC ACTATCCAGT GGAGGTGATG
     1561 CTGAAGGGGG GCGGACCCAA AAAGAAGCGC AAGGTTTGA
//
```

Fig. 19(C)

```
                9          18         27         36         45         54
5'  ATG GGA TGG AGC TGT ATC ATC CTC TTC TTG GTA GCA ACA GCT ACA GGT GTC CAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     M   G   W   S   C   I   I   L   F   L   V   A   T   A   T   G   V   H

               63         72         81         90         99        108
    TCC CAG GTG CAG CTG GTG CAG TCT GGG GCA GAG GTG AAA AAG CCT GGG GCC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     S   Q   V   Q   L   V   Q   S   G   A   E   V   K   K   P   G   A   S

              117        126        135        144        153        162
    GTG AAG GTG TCC TGC AAG GCT TCT GGC TAC ACC TTC AGT GCC TAC TGG ATA GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   K   V   S   C   K   A   S   G   Y   T   F   S   A   Y   W   I   E

              171        180        189        198        207        216
    TGG GTG CGC CAG GCT CCA GGA AAG GGC CTC GAG TGG GTC GGA GAG ATT TTA CCT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   V   R   Q   A   P   G   K   G   L   E   W   V   G   E   I   L   P

              225        234        243        252        261        270
    GGA AGT AAT AAT TCT AGA TAC AAT GAG AAG TTC AAG GGC CGA GTG ACA GTC ACT
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   S   N   N   S   R   Y   N   E   K   F   K   G   R   V   T   V   T

              279        288        297        306        315        324
    AGA GAC ACA TCC ACA AAC ACA GCC TAC ATG GAG CTC AGC AGC CTG AGG TCT GAG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     R   D   T   S   T   N   T   A   Y   M   E   L   S   S   L   R   S   E

              333        342        351        360        369        378
    GAC ACA GCC GTC TAT TAC TGT GCA AGA TCC TAC GAC TTT GCC TGG TTT GCT TAC
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     D   T   A   V   Y   Y   C   A   R   S   Y   D   F   A   W   F   A   Y

              387        396        405        414        423        432
    TGG GGC CAA GGG ACT CTG GTC ACA GTC TCC TCA GCC TCC ACC AAG GGC CCA TCG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     W   G   Q   G   T   L   V   T   V   S   S   A   S   T   K   G   P   S

              441        450        459        468        477        486
    GTC TTC CCC CTG GCA CCC TCC TCC AAG AGC ACC TCT GGG GGC ACA GCG GCC CTG
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     V   F   P   L   A   P   S   S   K   S   T   S   G   G   T   A   A   L

              495        504        513        522        531        540
    GGC TGC CTG GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   C   L   V   K   D   Y   F   P   E   P   V   T   V   S   W   N   S

              549        558        567        576        585        594
    GGC GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA GGA
    --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
     G   A   L   T   S   G   V   H   T   F   P   A   V   L   Q   S   S   G
```


Fig. 19(D)
(Sheet 1 of 3)

```
        603         612         621         630         639         648
CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG GGC ACC CAG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 L   Y   S   L   S   S   V   V   T   V   P   S   S   S   L   G   T   Q

        657         666         675         684         693         702
ACC TAC ATC TGC AAC GTG AAT CAC AAG CCC AGC AAC ACC AAG GTG GAC AAG AAA
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 T   Y   I   C   N   V   N   H   K   P   S   N   T   K   V   D   K   K

        711         720         729         738         747         756
GTT GAG CCC AAA TCT TGT GAC AAA ACT CAC ACA TGC TGT GTG GAG TGC CCA CCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   E   P   K   S   C   D   K   T   H   T   C   C   V   E   C   P   P

        765         774         783         792         801         810
TGC CCA GCA CCT GAA GGC GGG CTG AAG ATC GCA GCC TTC AAC ATC CAG ACA TTT
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 C   P   A   P   E   G   G   L   K   I   A   A   F   N   I   Q   T   F

        819         828         837         846         855         864
GGG GAG ACC AAG ATG TCC AAT GCC ACC CTC GTC AGC TAC ATT GTG CAG ATC CTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 G   E   T   K   M   S   N   A   T   L   V   S   Y   I   V   Q   I   L

        873         882         891         900         909         918
AGC CGC TAC GAC ATC GCC CTG GTC CAG GAG GTC AGA GAC AGC CAC CTG ACT GCC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   R   Y   D   I   A   L   V   Q   E   V   R   D   S   H   L   T   A

        927         936         945         954         963         972
GTG GGG AAG CTG CTG GAC AAC CTC AAT CAG GAC GCA CCA GAC ACC TAT CAC TAC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   G   K   L   L   D   N   L   N   Q   D   A   P   D   T   Y   H   Y

        981         990         999        1008        1017        1026
GTG GTC AGT GAG CCA CTG GGA CGG AAC AGC TAT AAG GAG CGC TAC CTG TTC GTG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 V   V   S   E   P   L   G   R   N   S   Y   K   E   R   Y   L   F   V

       1035        1044        1053        1062        1071        1080
TAC AGG CCT GAC CAG GTG TCT GCG GTG GAC AGC TAC TAC TAC GAT GAT GGC TGC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 Y   R   P   D   Q   V   S   A   V   D   S   Y   Y   Y   D   D   G   C

       1089        1098        1107        1116        1125        1134
GAG CCC TGC GGG AAC GAC ACC TTC AAC CGA GAG CCA GCC ATT GTC AGG TTC TTC
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 E   P   C   G   N   D   T   F   N   R   E   P   A   I   V   R   F   F

       1143        1152        1161        1170        1179        1188
TCC CGG TTC ACA GAG GTC AGG GAG TTT GCC ATT GTT CCC CTG CAT GCG GCC CCG
--- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- --- ---
 S   R   F   T   E   V   R   E   F   A   I   V   P   L   H   A   A   P

       1197        1206        1215        1224        1233        1242
```

Fig. 19(D)

(Sheet 2 of 3)

```
GGG GAC GCA GTA GCC GAG ATC GAC GCT CTC TAT GAC GTC TAC CTG GAT GTC CAA
 G   D   A   V   A   E   I   D   A   L   Y   D   V   Y   L   D   V   Q

      1251        1260        1269        1278        1287        1296
GAG AAA TGG GGC TTG GAG GAC GTC ATG TTG ATG GGC GAC TTC AAT GCG GGC TGC
 E   K   W   G   L   E   D   V   M   L   M   G   D   F   N   A   G   C

      1305        1314        1323        1332        1341        1350
AGC TAT GTG AGA CCC TCC CAG TGG TCA TCC ATC CGC CTG TGG ACA AGC CCC ACC
 S   Y   V   R   P   S   Q   W   S   S   I   R   L   W   T   S   P   T

      1359        1368        1377        1386        1395        1404
TTC CAG TGG CTG ATC CCC GAC AGC GCT GAC ACC ACA GCT ACA CCC ACG CAC TGT
 F   Q   W   L   I   P   D   S   A   D   T   T   A   T   P   T   H   C

      1413        1422        1431        1440        1449        1458
GCC TAT GAC AGG ATC GTG GTT GCA GGG ATG CTG CTC CGA GGG GCC GTT GTT CCC
 A   Y   D   R   I   V   V   A   G   M   L   L   R   G   A   V   V   P

      1467        1476        1485        1494        1503        1512
GAC TCG GCT CTT CCC TTT AAC TTC CAG GCT GCC TAT GGC CTG AGT GAC CAA CTG
 D   S   A   L   P   F   N   F   Q   A   A   Y   G   L   S   D   Q   L

      1521        1530        1539        1548        1557        1566
GCC CAA GCC ATC AGT GAC CAC TAT CCA GTG GAG GTG ATG CTG AAG GGG GGC GGA
 A   Q   A   I   S   D   H   Y   P   V   E   V   M   L   K   G   G   G

      1575        1584
CCC AAA AAG AAG CGC AAG GTT TGA 3'
 P   K   K   K   R   K   V   *
```

Fig. 19(D)
(Sheet 3 of 3)

Mammalian expression of humanised HMFG1-D Nase constructs

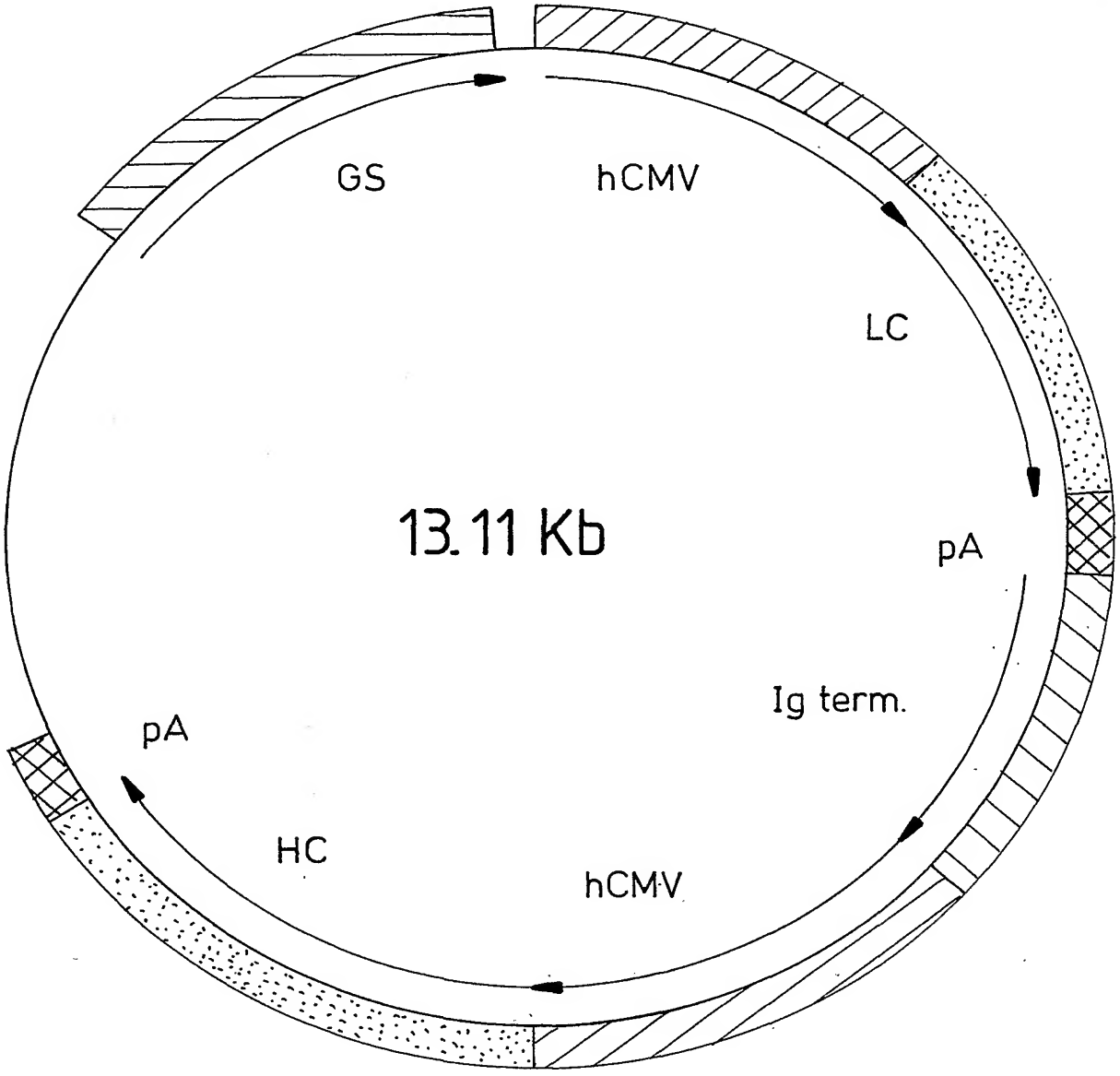


Fig. 20

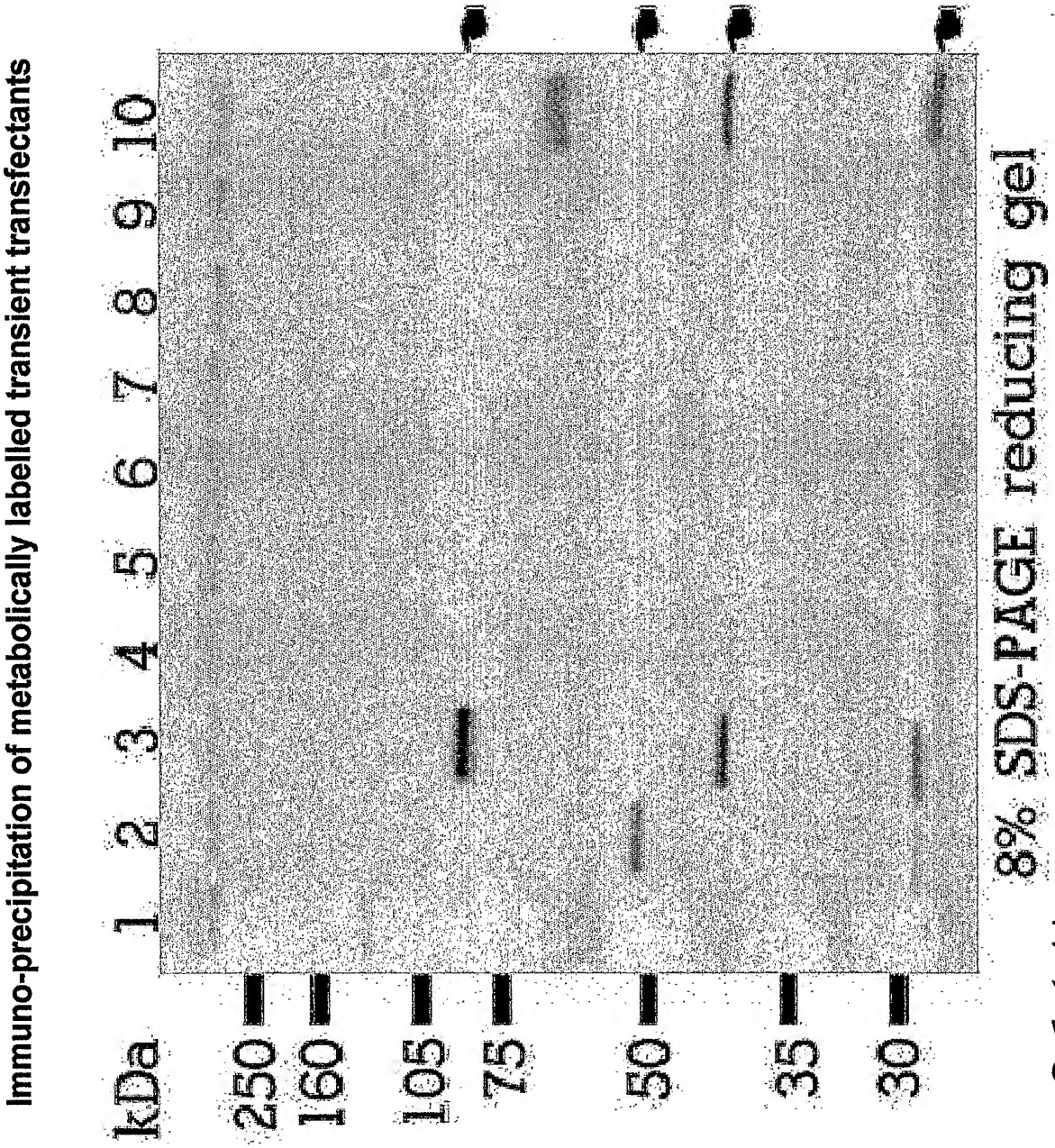

Immuno-precipitation of metabolically labelled transient transfectants
kDa
250
160
105
75
50
35
30
1 2 3 4 5 6 7 8 9 10
8% SDS-PAGE reducing gel

Fig. 21(A)

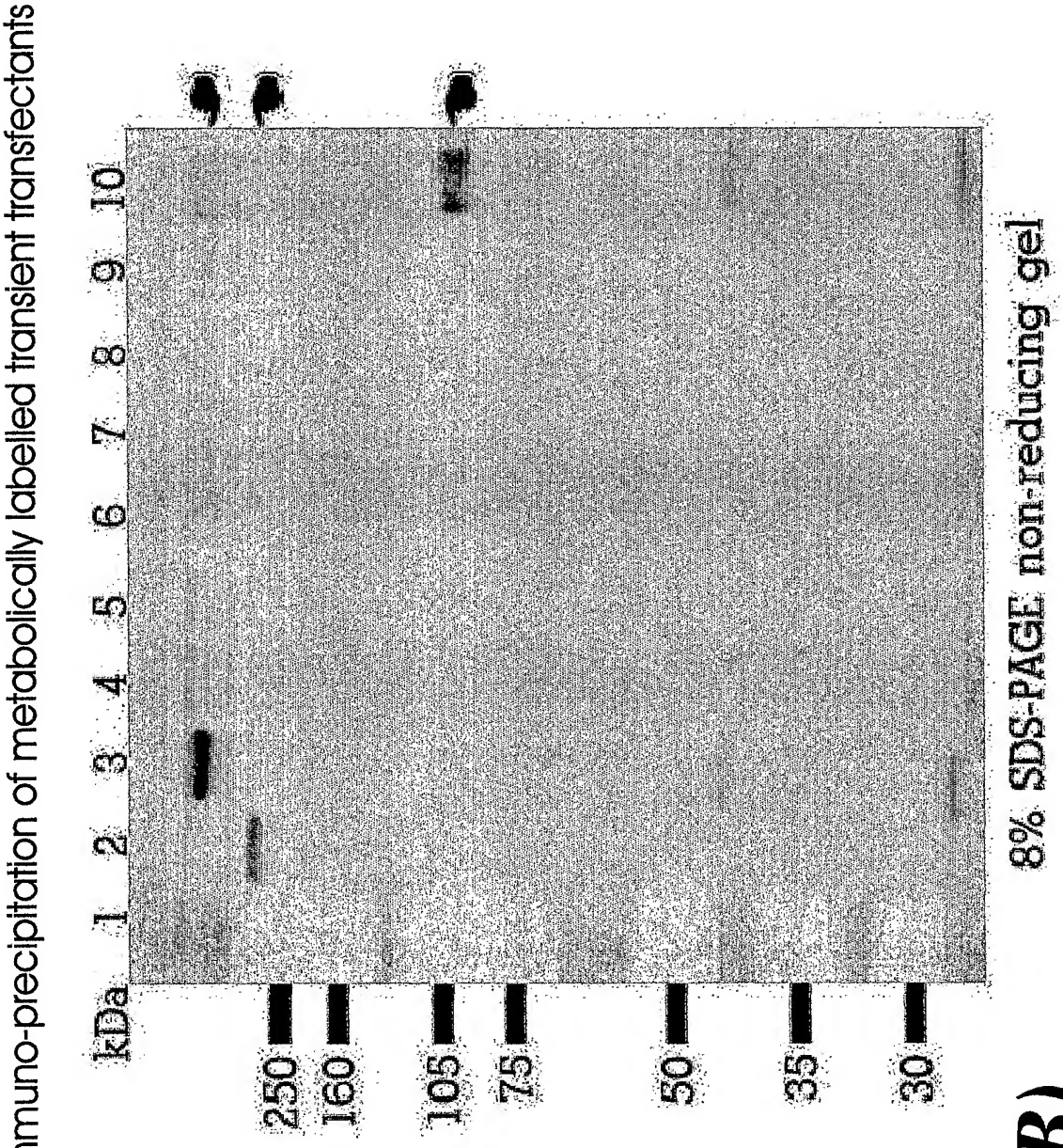

Immuno-precipitation of metabolically labelled transient transfectants
kDa
1
2
3
4
5
6
7
8
9
10
250
160
105
75
50
35
30
8% SDS-PAGE non-reducing gel

Fig. 21(B)

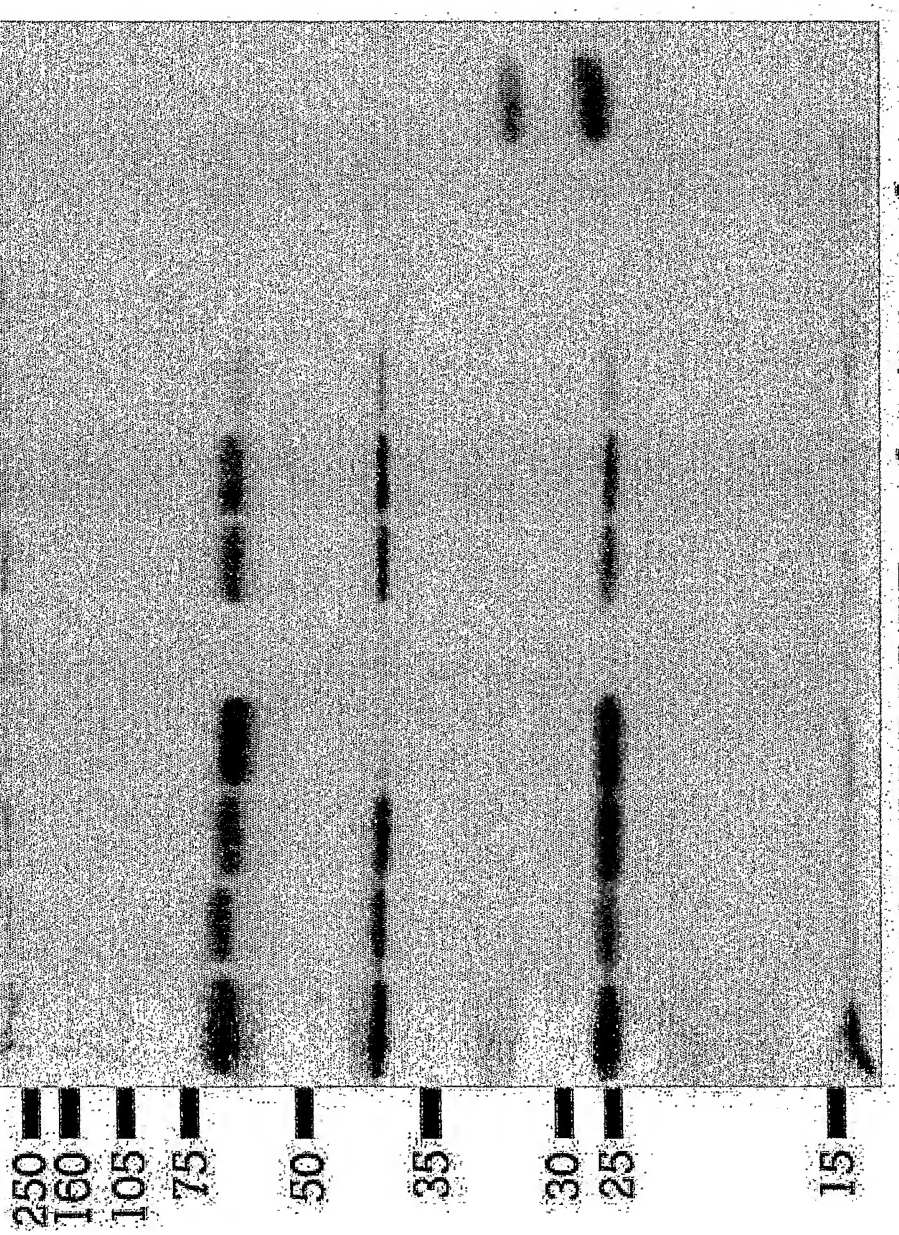

Immuno-precipitation of metabolically labelled transient transfectants
1 2 3 4 5 6 7 8 9 10 11
250
160
105
75
50
35
30
25
15
10% SDS-PAGE reducing gel

Fig. 21(C)

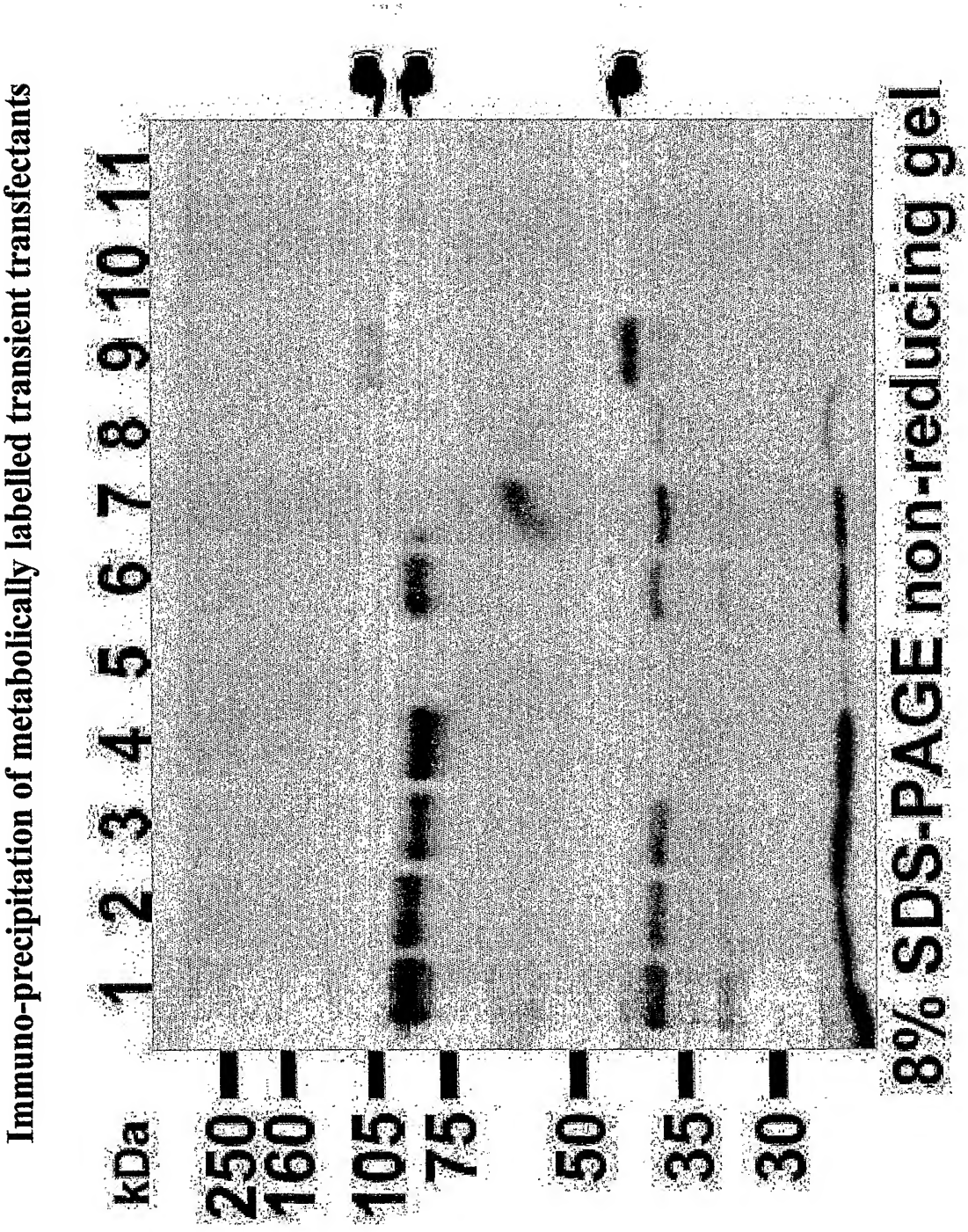

Immuno-precipitation of metabolically labelled transient transfectants
1 2 3 4 5 6 7 8 9 10 11
kDa
250
160
105
75
50
35
30
8% SDS-PAGE non-reducing gel

Fig. 21(D)

Fig. 22

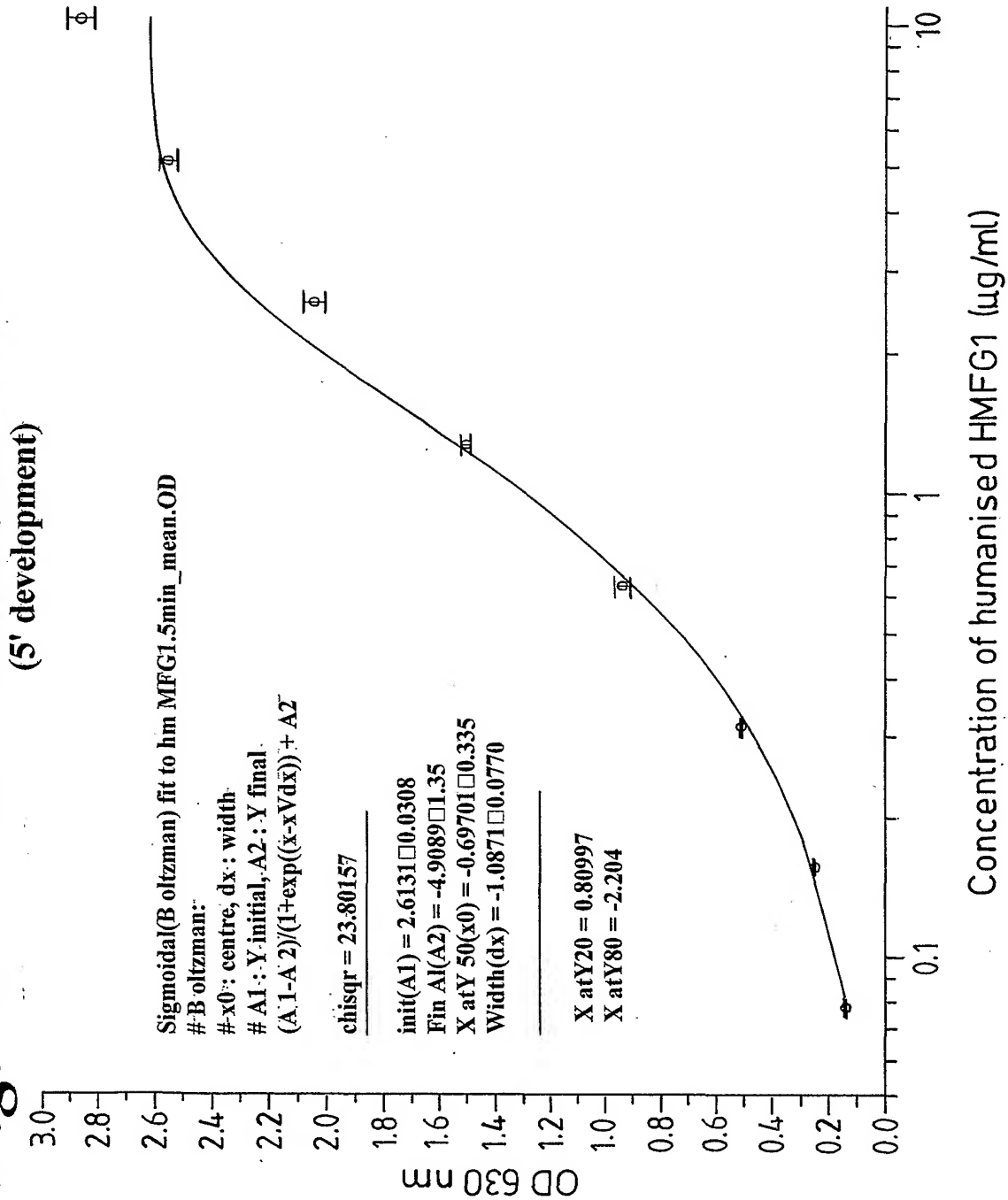
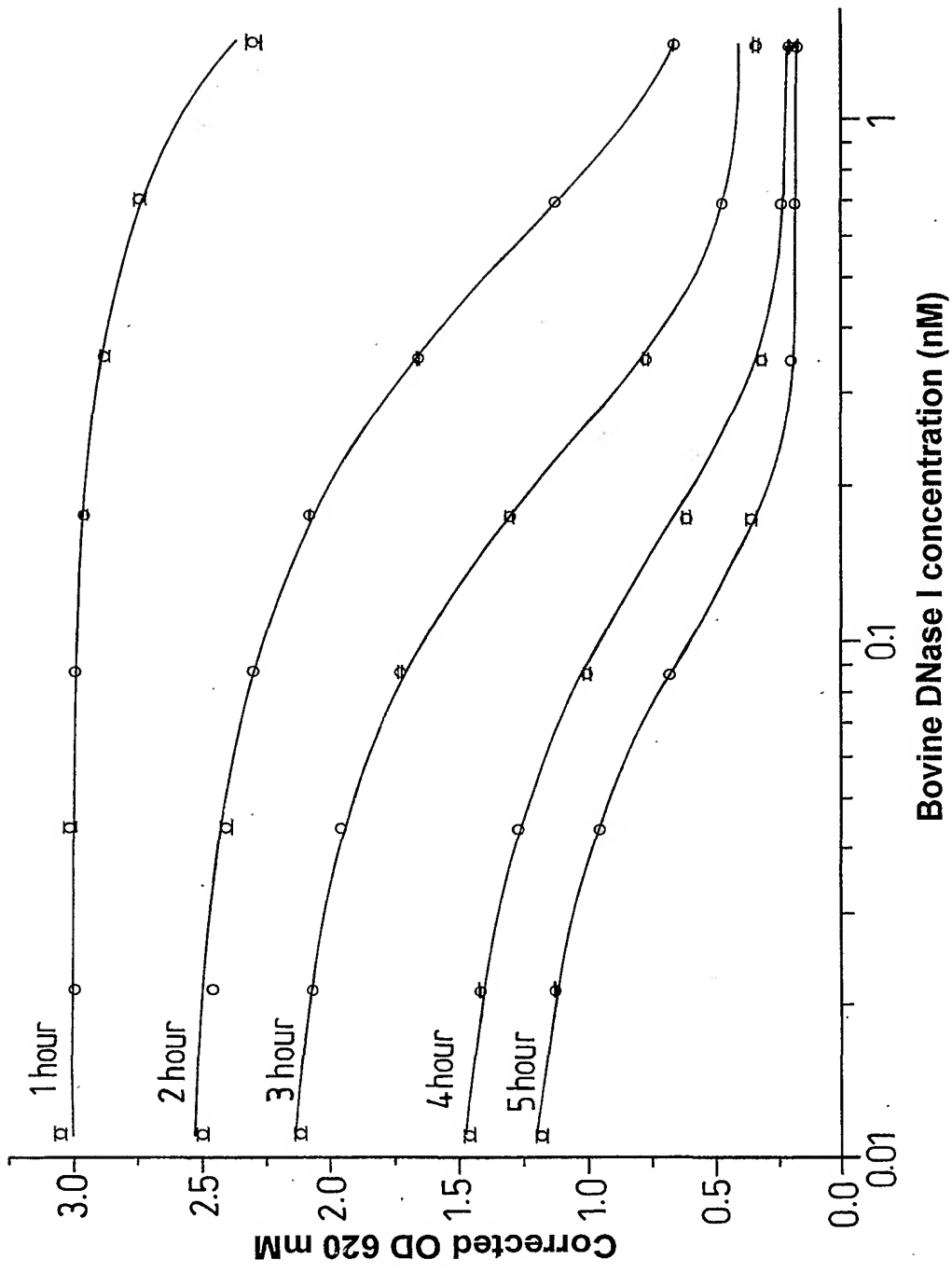

PDTRP binding assay standard curve
(5' development)
Sigmoidal(B oltzman) fit to hm MFG1.5min_mean.OD
#B oltzman:
#x0: centre, dx: width
A1: Y initial, A2: Y final
(A1-A2)/(1+exp((x-x0)/dx))+ A2
chisqr = 23.80157
init(A1) = 2.6131□0.0308
Fin Al(A2) = -4.9089□1.35
X atY 50(x0) = -0.69701□0.335
Width(dx) = -1.0871□0.0770
X atY20 = 0.80997
X atY80 = -2.204
OD 630 nm
3.0
2.8
2.6
2.4
2.2
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
0.1
1
10
Concentration of humanised HMFG1 (ug/ml)

Fig. 23


Corrected bovine DNase I standard curves
at various time points
Corrected OD 620 mM
Bovine DNase I concentration (nM)
1 hour
2 hour
3 hour
4 hour
5 hour
3.0
2.5
2.0
1.5
1.0
0.5
0.0
0.01
0.1
1

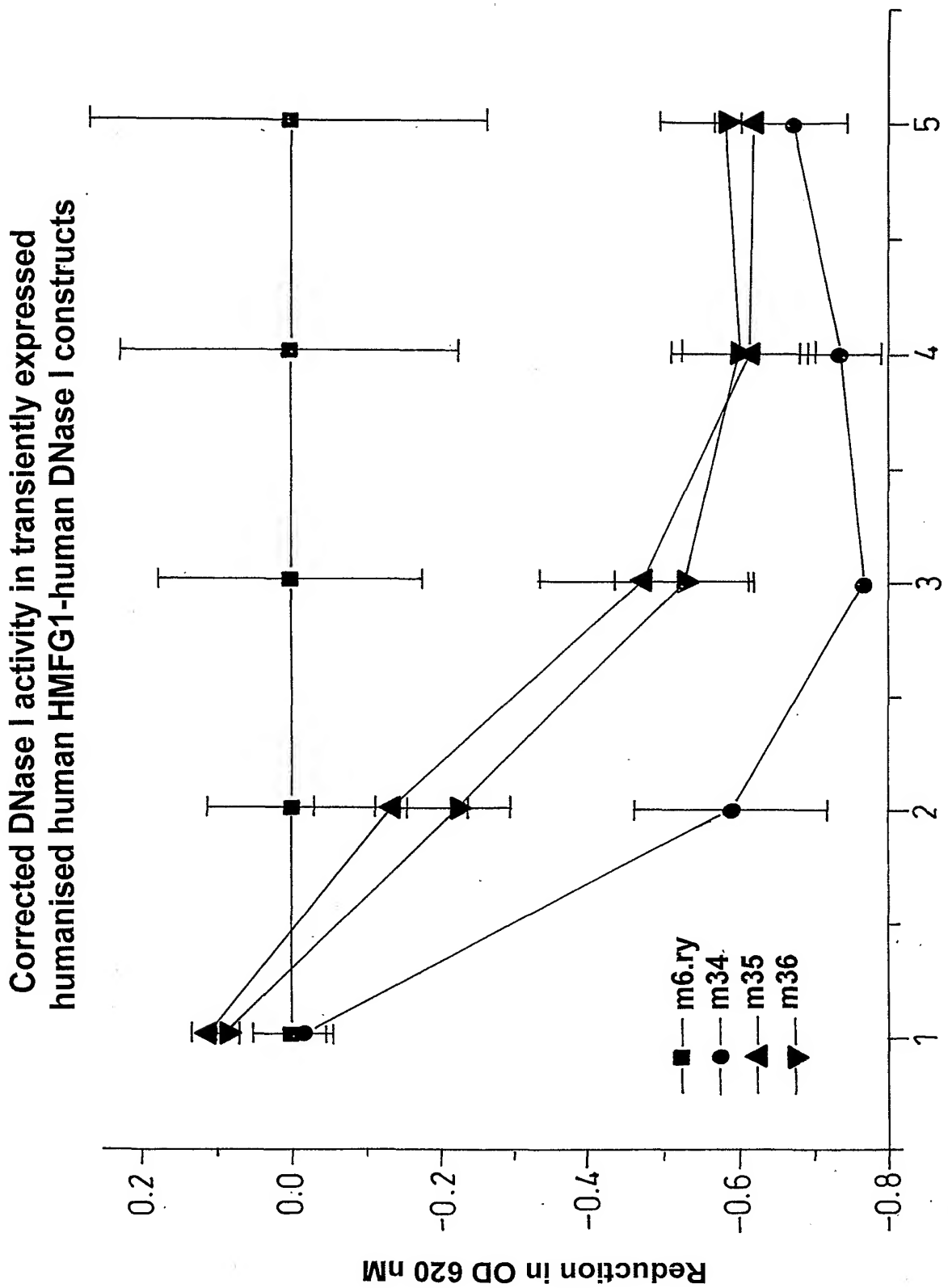

Corrected DNase I activity in transiently expressed humanised human HMFG1-human DNase I constructs
Reduction in OD 620 nM
0.2
0.0
-0.2
-0.4
-0.6
-0.8
1
2
3
4
5
Elapsed time (h)
m6.ry
m34
m35
m36

Fig. 24

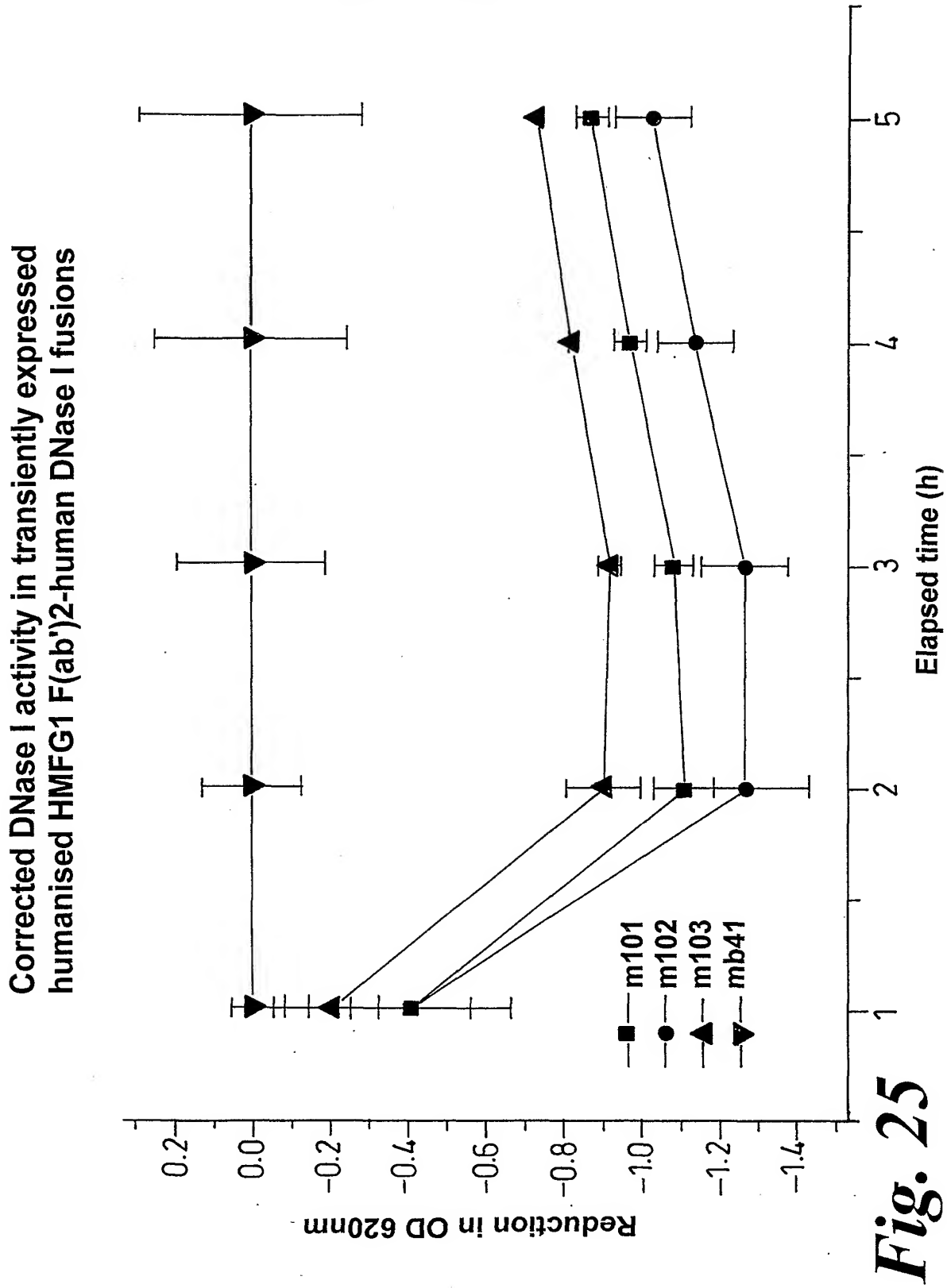

Corrected DNase I activity in transiently expressed humanised HMFG1 F(ab')2-human DNase I fusions
Reduction in OD 620nm
0.2
0.0
-0.2
-0.4
-0.6
-0.8
-1.0
-1.2
-1.4
1
2
3
4
5
Elapsed time (h)
m101
m102
m103
mb41

Fig. 25

Fig. 26

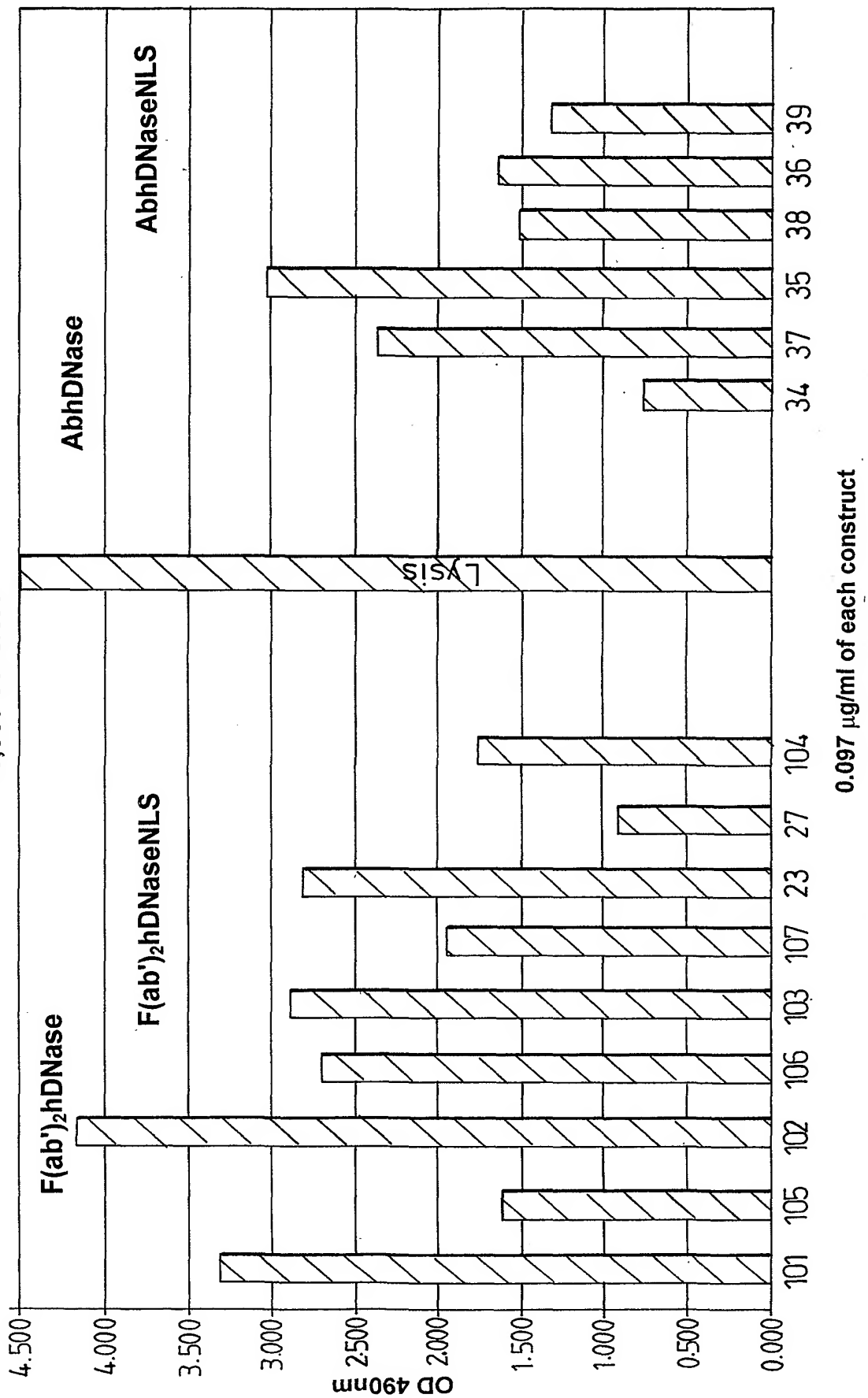
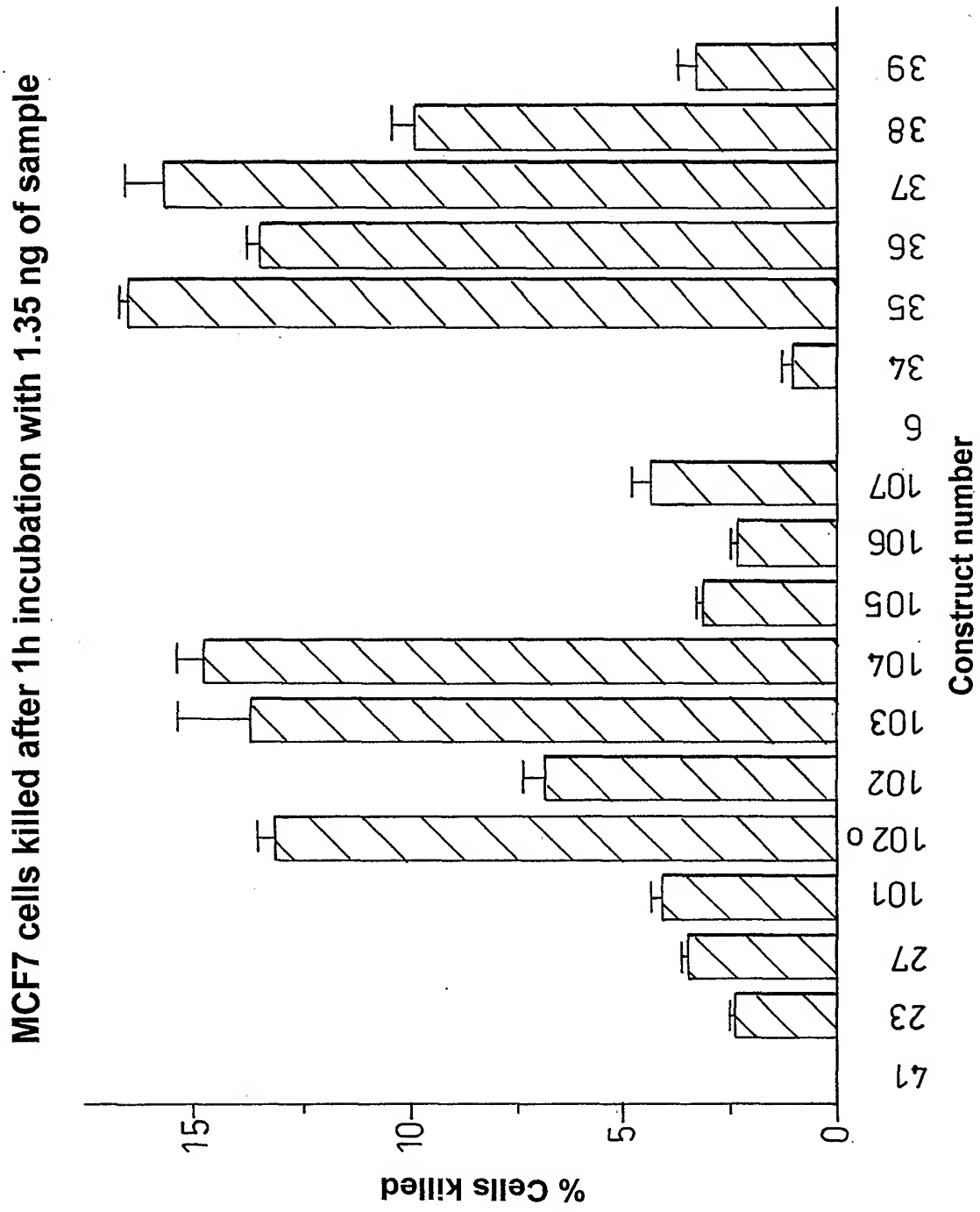

Cytoxicity Assay
10,000 cells/Well
F(ab')2hDNase
F(ab')2hDNaseNLS
LYSIS
AbhDNase
AbhDNaseNLS
OD 490nm
4.500
4.000
3.500
3.000
2500
2.000
1.500
1.000
0.500
0.000
101 105 102 106 103 107 23 27 104 34 37 35 38 36 39
0.097 µg/ml of each construct

Fig. 27


MCF7 cells killed after 1h incubation with 1.35 ng of sample
% Cells killed
0
5
10
15
41
23
27
101
102 o
102
103
104
105
106
107
6
34
35
36
37
38
39
Construct number

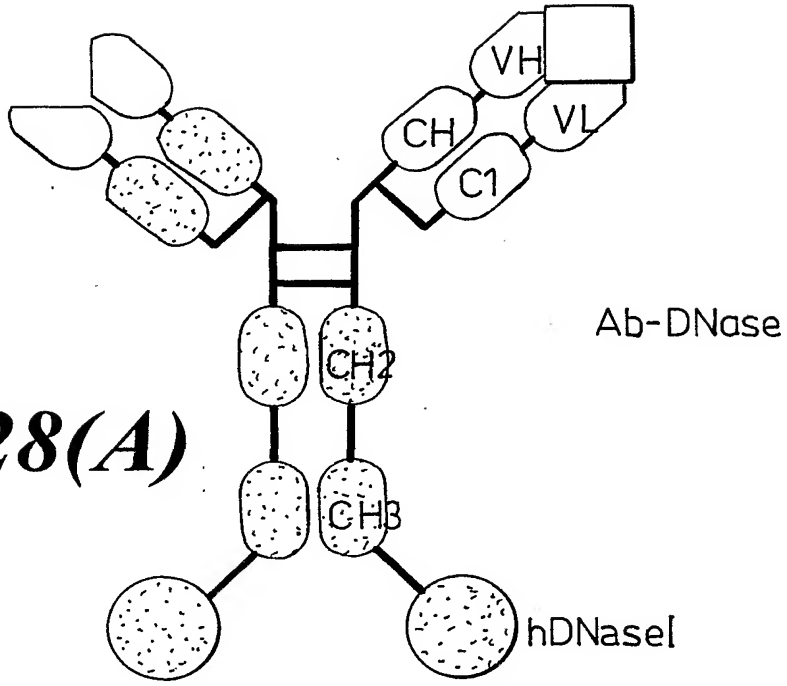


Fig. 28(A)

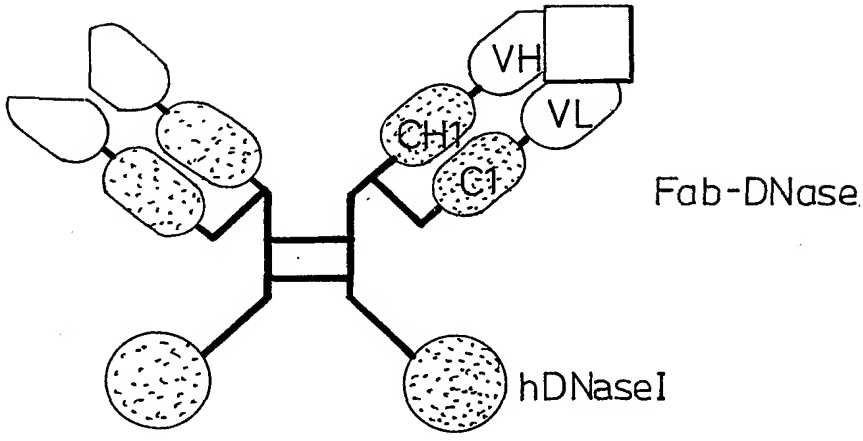
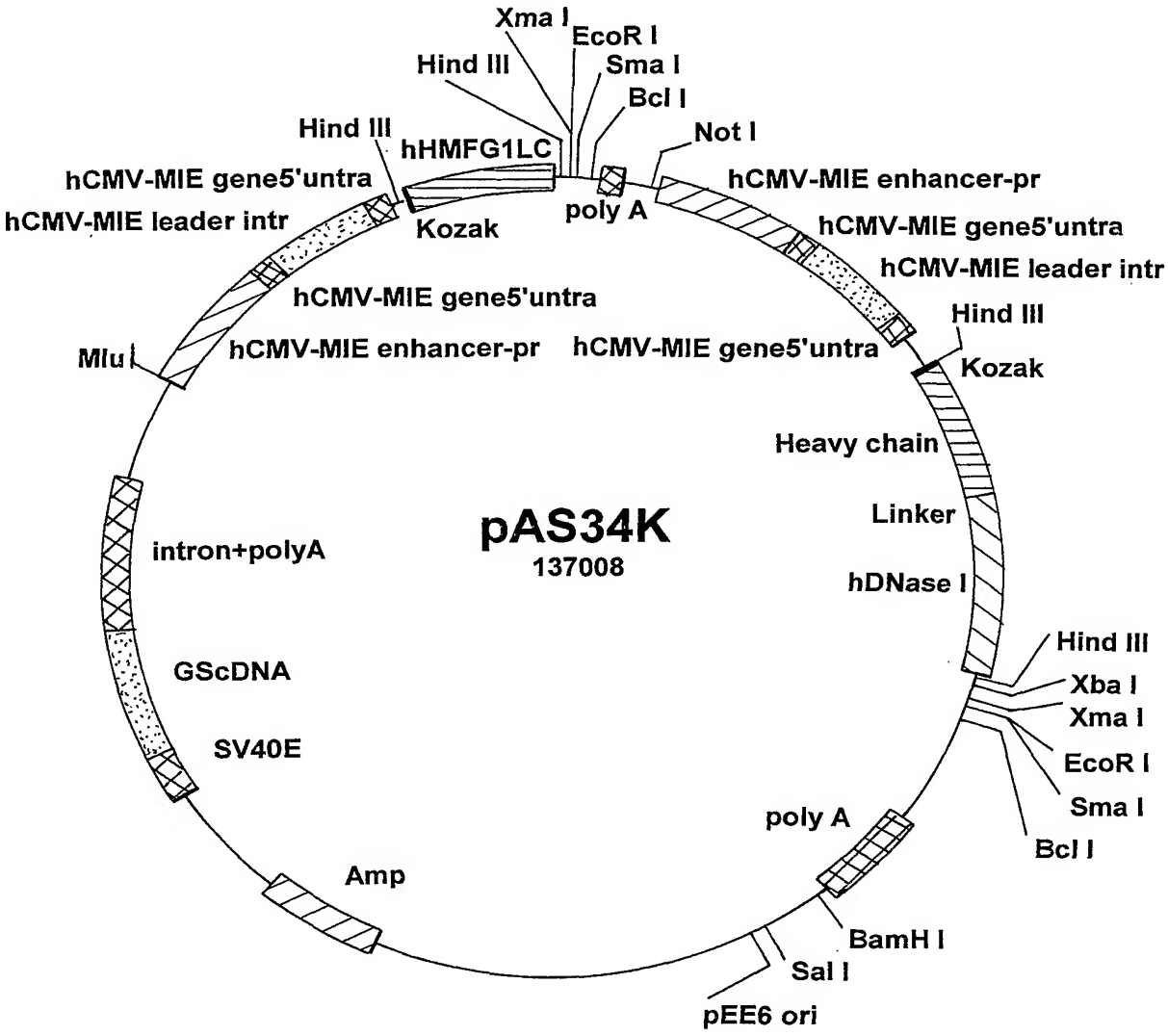
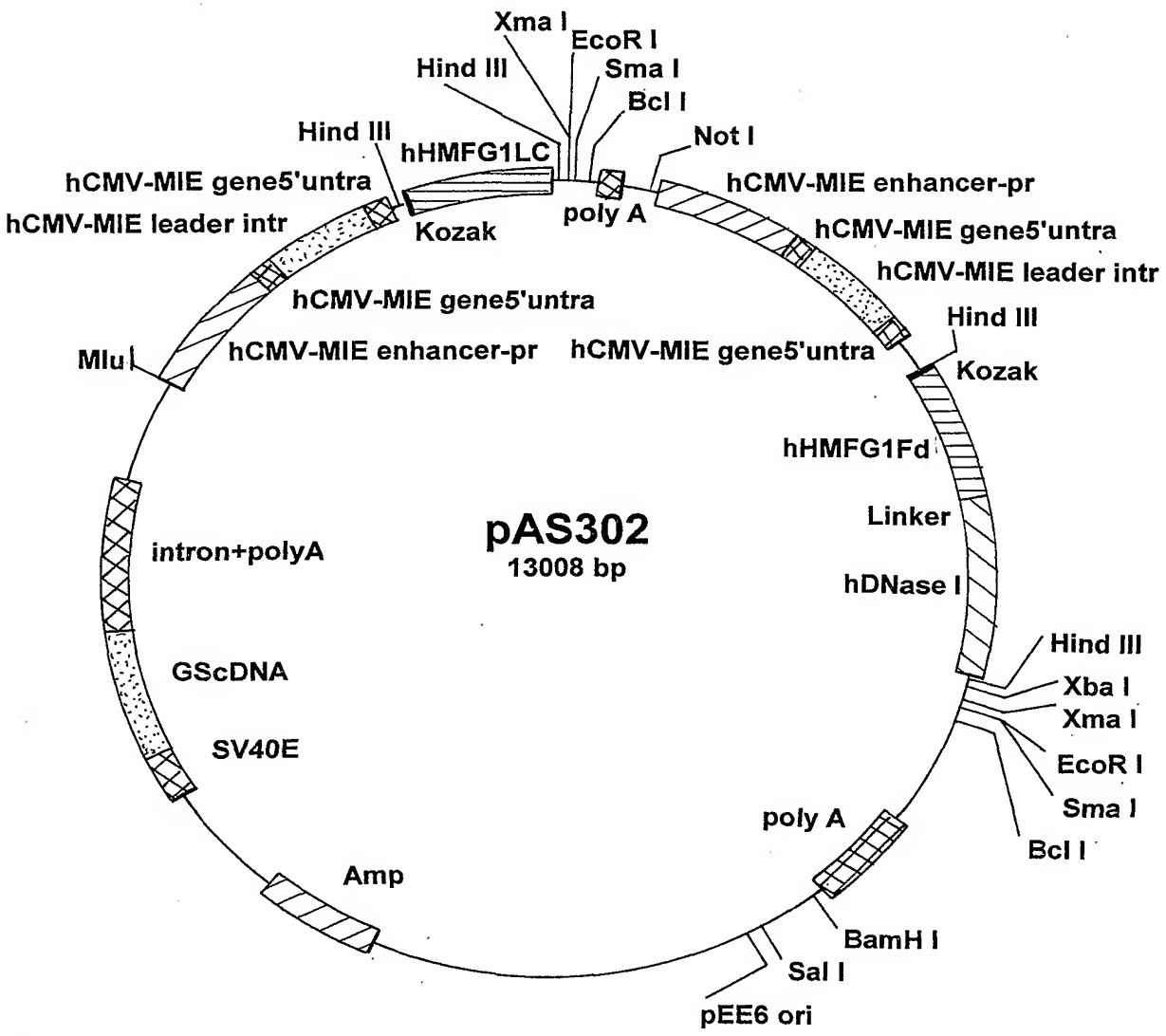


Fig. 28(B)



Ab-DNase

Fig. 29



Fab-DNase

Fig. 30

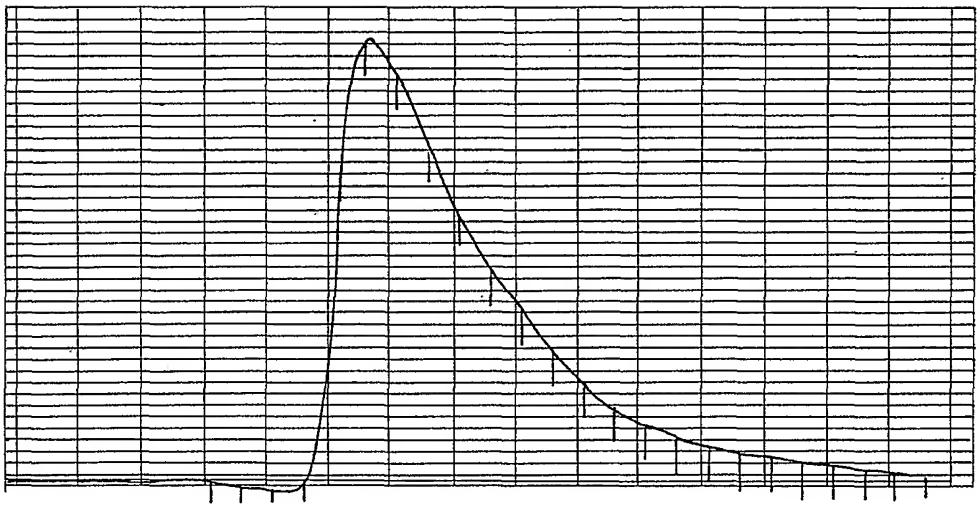


Fig. 31(A)

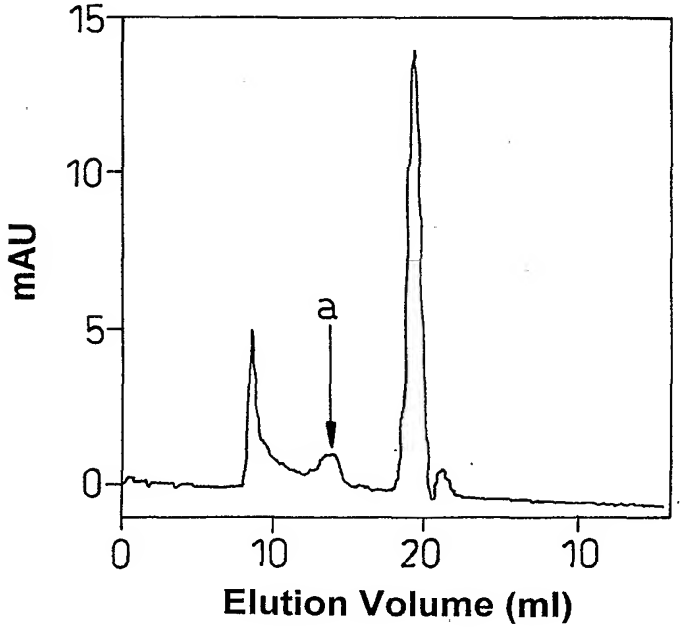


Fig. 31(B)

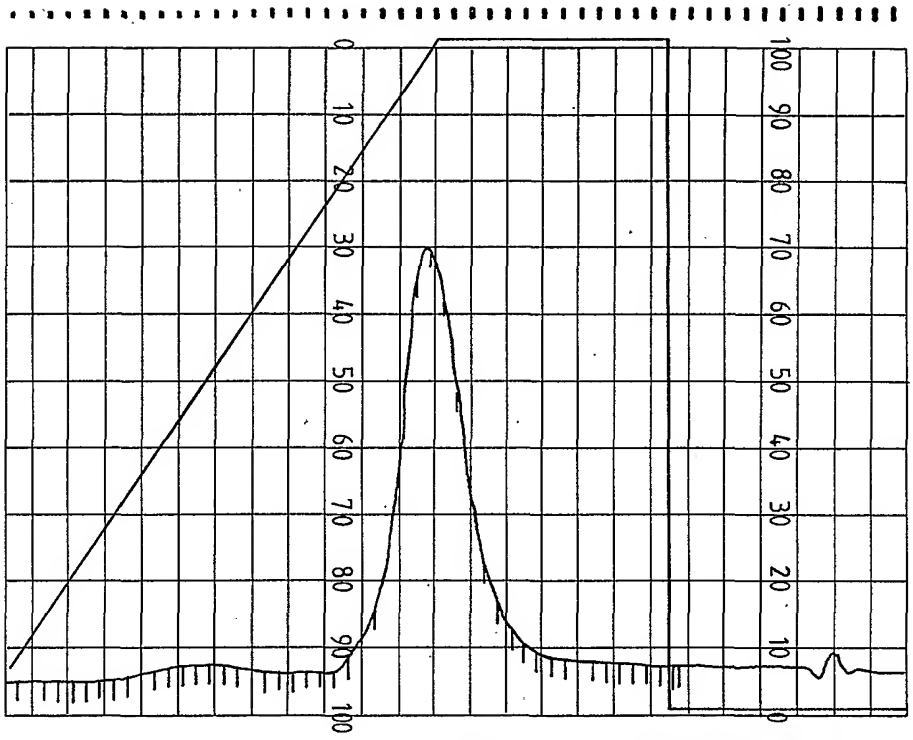


Fig. 32(A)

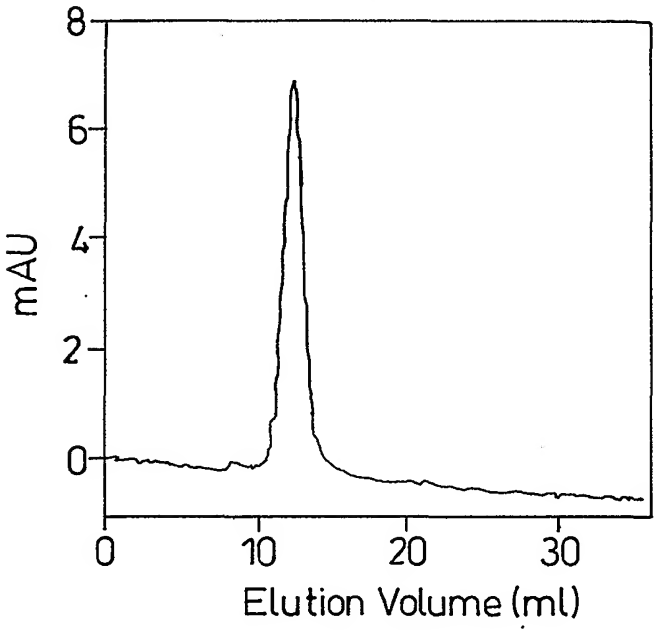


Fig. 32(B)

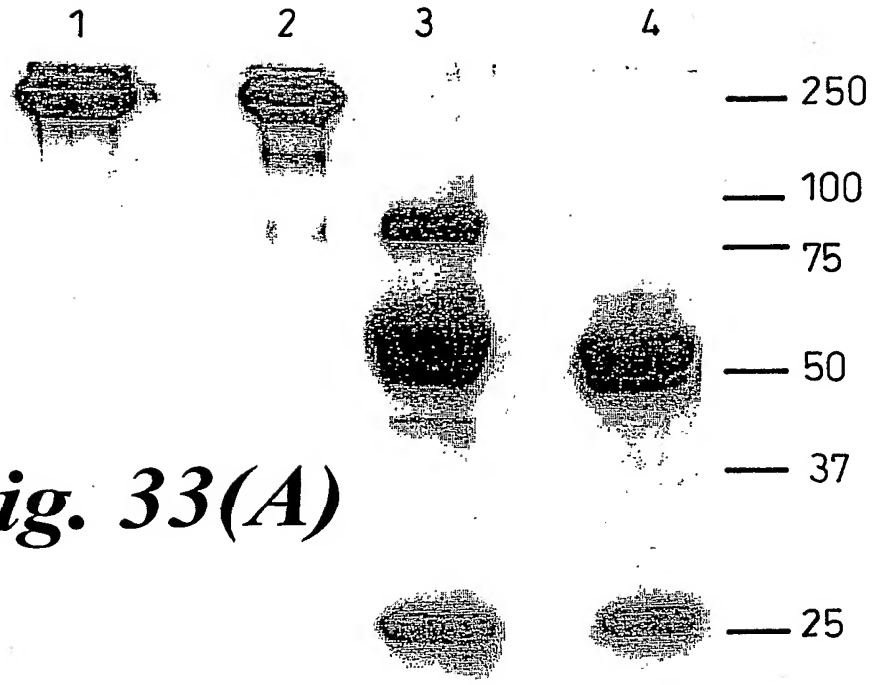


Fig. 33(A)

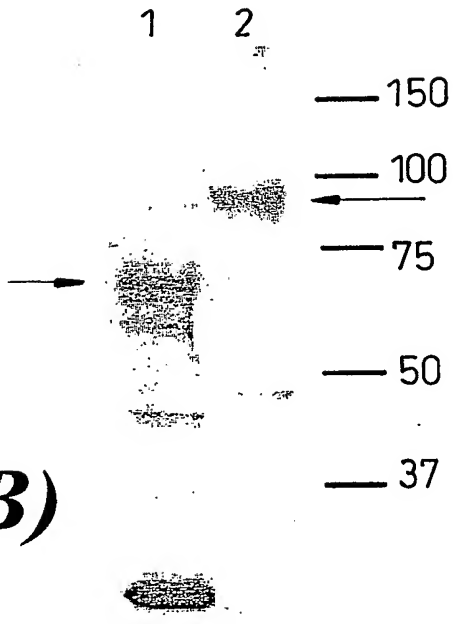


Fig. 33(B)

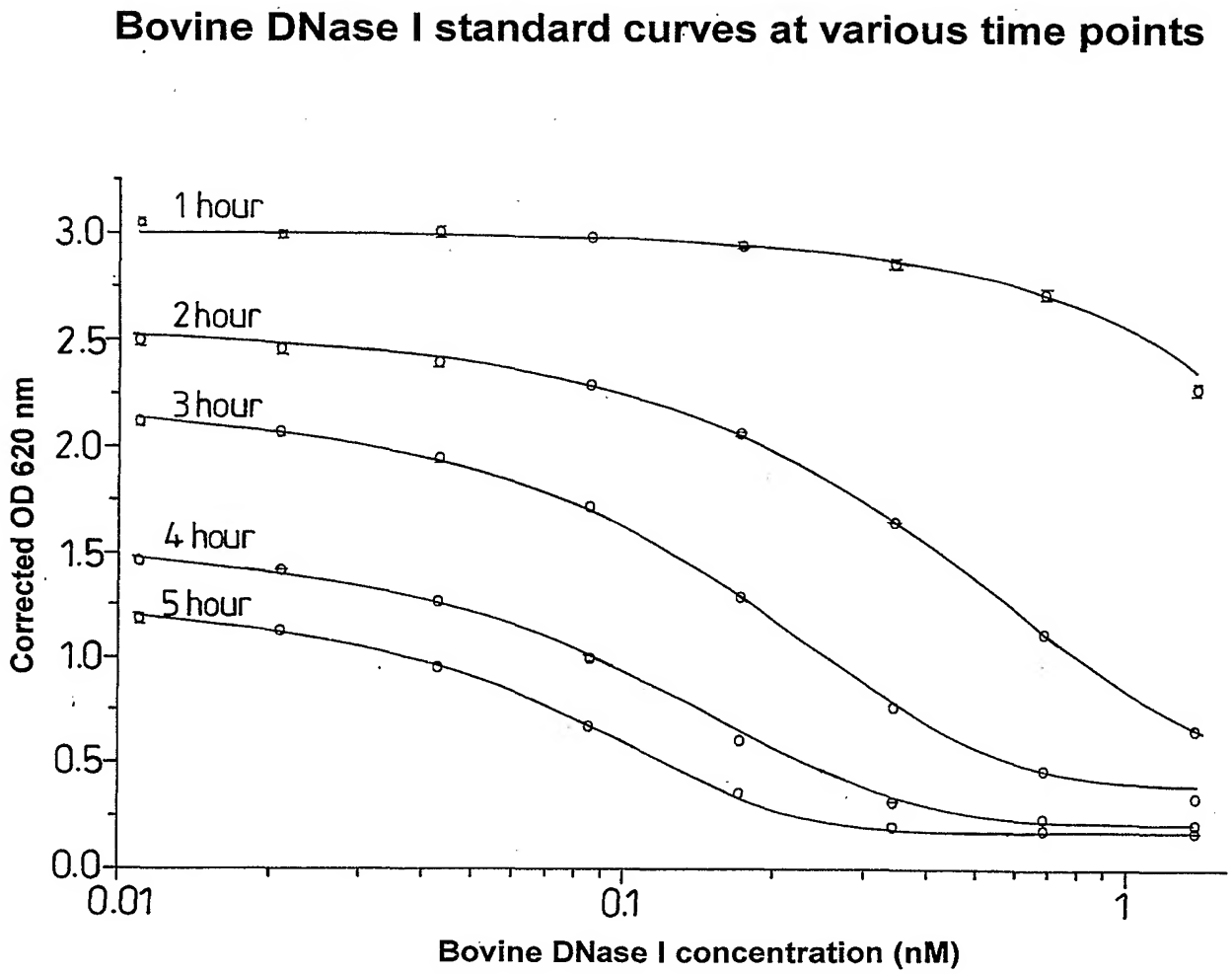

Bovine DNase I standard curves at various time points
1 hour
2 hour
3 hour
4 hour
5 hour
Corrected OD 620 nm
3.0
2.5
2.0
1.5
1.0
0.5
0.0
0.01
0.1
1
Bovine DNase I concentration (nM)

Fig. 34(A)

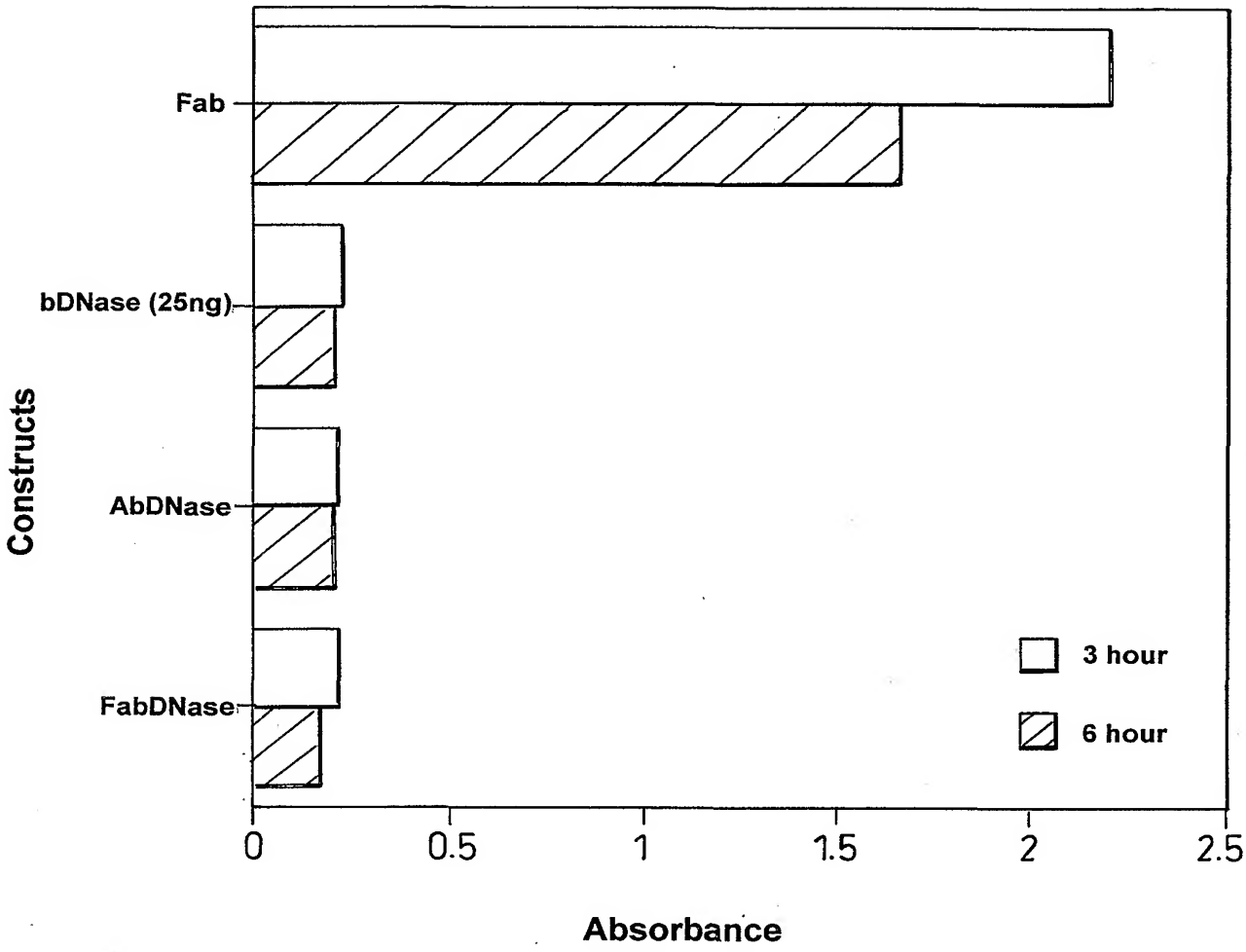

Fab
bDNase (25ng)
AbDNase
FabDNase
Constructs
3 hour
6 hour
0
0.5
1
1.5
2
2.5
Absorbance

Fig. 34(B)

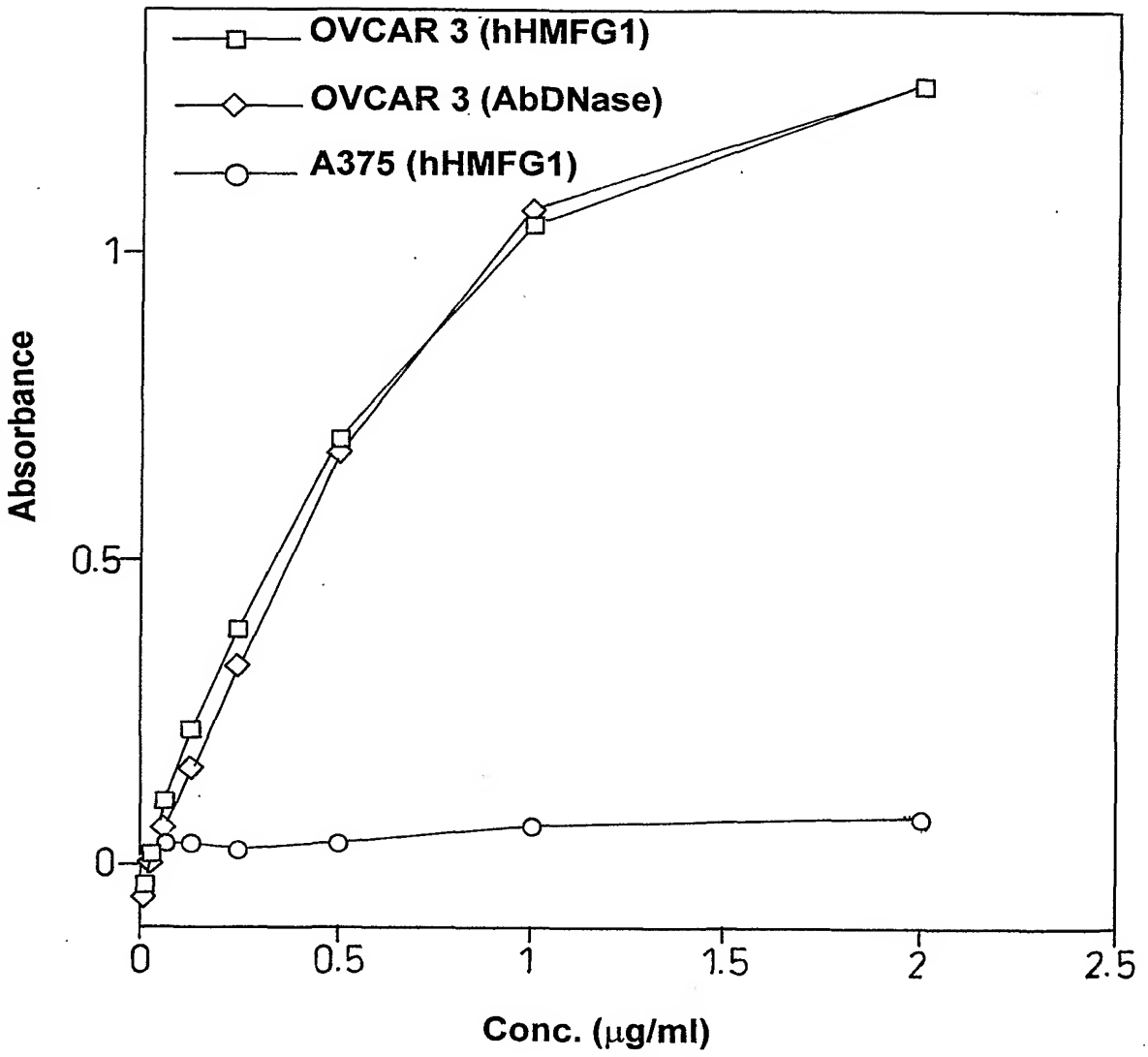

OVCAR 3 (hHMFG1)
OVCAR 3 (AbDNase)
A375 (hHMFG1)
Absorbance
0
0.5
1
0
0.5
1
1.5
2
2.5
Conc. (µg/ml)

Fig. 35(A)

113/113

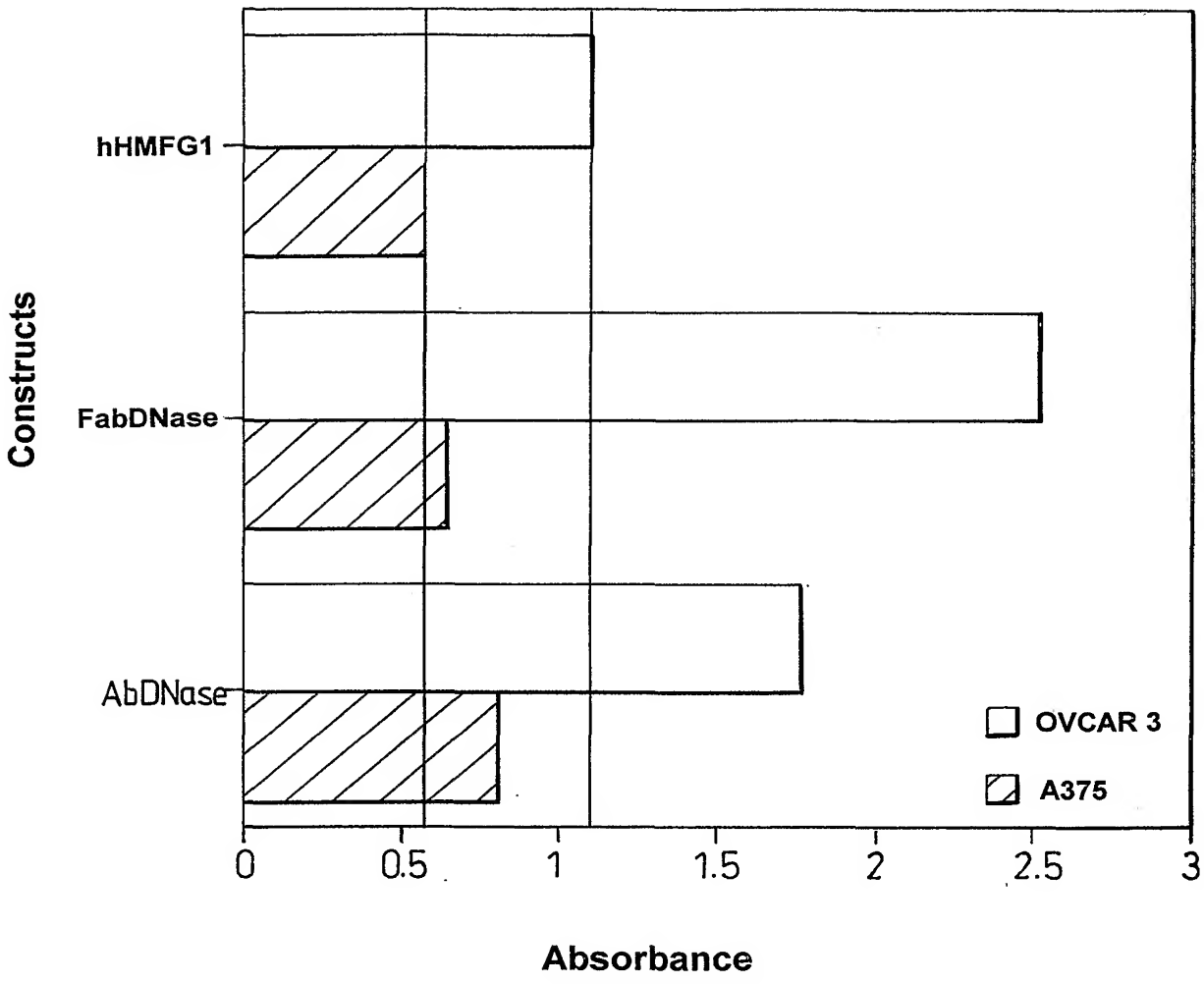


Fig. 35(B)

INTERNATIONAL SEARCH REPORT

ational Application No
PCT/GB 01/01324

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C07K16/18 C12N9/22 C12N15/62 C07K16/46 C12N15/63
C12N15/85 A61K39/395 A61K38/43 //C07K19/00

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 C07K C12N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)
BIOSIS, EPO-Internal, WPI Data, MEDLINE, PAJ

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	YOUNG ROBERT J ET AL: "A DNase I based immunotoxin for tumor therapy." PROCEEDINGS OF THE AMERICAN ASSOCIATION FOR CANCER RESEARCH ANNUAL, no. 41, March 2000 (2000-03), page 289 XP001008862 91st Annual Meeting of the American Association for Cancer Research.;San Francisco, California, USA; April 01-05, 2000, March, 2000 ISSN: 0197-016X abstract --- -/--	1-12,14, 15,17, 21,23, 25, 28-35, 37,38

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
6 August 2001	16/08/2001

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL – 2280 HV Rijswijk Tel. (+31–70) 340–2040, Tx. 31 651 epo nl, Fax: (+31–70) 340–3016	Montrone, M

Form PCT/ISA/210 (second sheet) (July 1992)

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication,where appropriate, of the relevant passages	Relevant to claim No.
X	WO 94 15644 A (EPENETOS AGAMEMNON ANTONIOU ;IMP CANCER RES TECH (GB); DEONARAIN M) 21 July 1994 (1994-07-21) abstract page 3, line 12-22 page 4, line 24-26 page 6, line 7 -page 8, line 2 page 10, line 11-14 page 12, line 18 -page 13, line 8 page 15, line 1-19 page 26, line 30 -page 27, line 25 page 28, line 23 -page 29, line 21 page 49, line 29 -page 51, line 10	1-6, 9-19,23, 28-38
Y	LINARDOU H. ET AL.: "Deoxyribonuclease I (DNase I). A novel approach for targeted cancer therapy." CELL BIOPHYS., vol. 24-25, 1994, page 243-248 XP001012902 abstract page 244, paragraphs 2-4 page 245, paragraph 4 page 246; figure 1 page 247, paragraphs 2,3,5	15,16, 18,19
Y	WO 92 04380 A (UNILEVER PLC ;UNILEVER NV (NL)) 19 March 1992 (1992-03-19) cited in the application abstract page 4, line 30 - line 27 page 6, line 6-26 page 8, line 30 -page 9, line 7 page 9, line 11-28 page 10, line 1-6 page 10, line 33 -page 11, line 3 page 12, line 13-35 page 14, line 14-16 page 15, line 15-30 page 16, line 15-20 page 17, line 6-14	1-19,21, 23,25, 27-38
Y	EP 0 781 845 A (CELLTECH THERAPEUTICS LTD) 2 July 1997 (1997-07-02) abstract page 3, line 19-48 page 5, line 3 -page 6, line 15 page 9, line 23-42 page 16, line 8-17	1-19,21, 23,25, 27-38

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.2

Claims Nos.: 20,22,24,26,38

Present claims 20, 24 and 38 relate to an extremely large number of possible compounds. In fact, the claims contain so many options, variables, possible permutations and provisos that a lack of clarity within the meaning of Article 6 PCT arises to such an extent as to render a meaningful search of the claims impossible. Moreover, a search for the subject-matter of claims 22 and 26 has not been carried out since it was not possible to identify the corresponding SEQ.ID.NO. of fig. 14(c). Consequently, the search has been carried out for those parts of the application which do appear to be clear, namely 1 to 19, 21, 23, 25, 27 to 37.

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

INTERNATIONAL SEARCH REPORT

Information on patent family members

ational Application No
PCT/GB 01/01324

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9415644 A	21-07-1994	AT 164314 T	15-04-1998
		DE 69409225 D	30-04-1998
		DE 69409225 T	13-08-1998
		DK 679094 T	19-10-1998
		EP 0679094 A	02-11-1995
		EP 0815872 A	07-01-1998
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		GB 2289679 A,B	29-11-1995
		GB 2300859 A,B	20-11-1996
		GR 3026449 T	30-06-1998
		JP 8509460 T	08-10-1996
		US 5973116 A	26-10-1999
WO 9204380 A	19-03-1992	AU 653167 B	22-09-1994
		AU 8495391 A	30-03-1992
		BG 60716 B	31-01-1996
		BG 97607 A	31-03-1994
		CA 2090961 A	08-03-1992
		EP 0483961 A	06-05-1992
		FI 930984 A	05-03-1993
		HU 67796 A	28-04-1995
		JP 6500468 T	20-01-1994
		NO 930825 A	05-05-1993
		RO 113432 B	30-07-1998
		US 6204366 B	20-03-2001
EP 0781845 A	02-07-1997	AT 160362 T	15-12-1997
		AU 666868 B	29-02-1996
		AU 2598392 A	27-04-1993
		CA 2095926 A	27-03-1993
		DE 69223206 D	02-01-1998
		DE 69223206 T	25-06-1998
		EP 0534742 A	31-03-1993
		ES 2108732 T	01-01-1998
		WO 9306231 A	01-04-1993
		IL 103269 A	04-01-1998
		JP 6505399 T	23-06-1994
		NZ 244468 A	25-11-1994